RECENT ADVANCES IN MEDICINE

Clinical Laboratory Therapeutic

BY

G E BEAUMONT

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THIRTEENTH EDITION

With 59 Illustrations



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PREFACE TO THE THIRTEENTH EDITION

This edition contains about two hundred pages of new material and twenty nine new figures As the size of the book has not been increased it has been necessary to omit completely certain chapters together with sections of others The chapters we have discarded are those dealing with Chemotherapy Glycosuria and Diabetes Mellitus The Treatment of Addison s Disease and The Sex Hormones

New Chapters have been written on The Collagen Diseases The Antihistamines and The Use of Isotopes in Medicine The last Chapter on Biochemical Methods has been rewritten The Antibiotic Chapter has been revised and expanded to include an account of Aureomycin Chloramphenicol Streptomycin and Terramycin

Other additions include sections on Vitamin B_1 a general con sideration of hepatitis and liver puncture biopsy The description of the tests for liver function has been rewritten The medical treatment of gastric ulcer has been brought up to date and an article added on vagotomy In the Chapter dealing with The Cardiovascular System sections have been added on Unipolar Lead Electrocardiography Cardiac Catheterisation and the use of methonium salts and the rice diet in hypertension

Further accounts are given of Q Fever in Great Britain and the method which may be adopted for the prevention of tuber culosis including the B C G vaccine Artificial pneumoperitoneum and the streptomycin and P A S treatments of tuberculosis are also considered The section on Bronchography has been rewritten to include the various techniques which may be adopted for adults and children The description of the use of thiouracil drugs has been revised and a section added on radioactive iodine in the

diagnosis and treatment of thyrotoxicosis The Hæmopoietic System Chapter has been brought up to date and an account given of the intravenous administration of iron The section dealing with the blood groups has been completely rewritten

Medicine has advanced so rapidly during the past few years that it is difficult to keep apace but it is hoped that nothing of major importance has been omitted from this edition

We have received much help in its preparation from our colleagues and we take this opportunity of gratefully acknowledging our indebtedness Sir Weldon Dalrymple Champneys kindly supplied us with information on milk supplies Dr S Oram not only read the article on Unipolar Electrocardiography but also supplied all the electrocardiograms which illustrate this section For his valuable suggestions criticisms and amendments we are deeply grateful Dr I H Scadding rewrote the article on Bronchography and gave helpful information for the account of Cardiac Catheterisation Dr J M Gardiner also read the latter article and made various useful suggestions To Dr A M Jelliffe we are obliged for the two figures illustrating Liver Needle Biopsy and for assistance in describing the technique of the method

Dr D N Baron and Mr C F M Rose have played a major part in the revision of the biochemical sections of the book and have assisted with the sections on antihistamines and the liver and the methods respectively To Drs Johnstone Palmer and Tait we are much obliged for assistance with the sections on blood groups antibiotics and isotopes respectively

We also wish to acknowledge with thanks permission to reproduce the following figures and diagrams Fig 4 Dr Myron Prinzmetal and *The Journal of the American Medical Association* Fig 5 Dr Edith H Quimby and *Radiology* Fig 6 Dr C F Green Dr J F Tait Mr R Worsnop and Professor B W Windeyer and *The British Journal of Radiology* Fig 48 Mr R C Brock and *Thorax* Fig 50 Professor J P Peters and Dr Donald D Van Slyke and The Williams and Wilkins Company Fig 59 Dr Franklin C McLean and *The American Journal of the Medical Sciences*

Our publishers have once again given us every help in the production of this edition and we wish especially to thank Mr J Rivers for his ready assistance at all times To Mr J Shields we owe the index of this volume and indeed of all the previous editions

C E B
E C D

LONDON

PREFACE TO THE FIRST EDITION

During the last decade changes have taken place in medicine especially in the routine methods adopted in the clinical and laboratory investigation of disease and also in certain forms of treatment

This book has been compiled with the following objects to assist practitioners who have not had the opportunity of recent post graduate study to familiarise themselves with some of these advances to provide a reference book for those who are working for the higher examinations in medicine to give candidates studying for the primary examination for the Fellowship of the Royal College of Surgeons an account of the application of physiological and biochemical principles to medicine It is also hoped that it will prove of assistance to the laboratory worker in that the recent chemical methods are dealt with in detail It should also form a link between the wards of a hospital and the laboratories giving fuller details of methods which are alluded to in medical textbooks but often omitted from the handbooks on clinical methods The recent work of American and Continental authorities has also been incorporated

One of the chief difficulties has been to decide the actual scope of the contents which of necessity encroach upon the domains of theoretical medicine therapeutics biochemistry and bacteriology The guiding principle has been to confine the subject matter to a description of such methods of diagnosis and treatment as are used for medical patients in a general hospital and which can be justly termed recent advances in medicine

Great care has been taken to give a workable description of each procedure all techniques described have been performed personally by one or other of us and the accounts are taken from our notebooks Although these may differ slightly from the original descriptions the methods described have been used by us as a routine for some time and have given very satisfactory

results We have attempted to state the value of the results obtained by the various tests

The authors wish to express their indebtedness to the numerous writers whose works have been consulted and an endeavour has been made to acknowledge them by the list of references

We have pleasure in thanking Sir Thomas Lewis for permission to reproduce the electrocardiograms taken from his book entitled Clinical Electrocardiography Dr D E Bedford has supplied the polygraph tracings and has also read through the proofs of the chapter dealing with the heart We are grateful for his valuable suggestions and help Further we wish to express our thanks for the loan of certain blocks to Messrs Hawksley & Sons for the one illustrating the Jacquet polygraph to Messrs Down Bros for those illustrating the pneumothorax needles and to Professor Harris for the diagram of the electrocardiograph from Anrep and Harris Practical Physiology

In conclusion we wish to acknowledge the unfailing help which we have received from the publishers of this volume

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CONTENTS

Clinic they decided together to investigate the activity of certain steroidal substances isolated from the suprarenal cortex and identified by Kendall One of these was the now familiar compound 'E' which will be dealt with from the chemical point of view at a later stage of this chapter Hench (1949b) (4) made the astonishing observation that the administration of compound 'E' daily in 100 mg doses resulted in a dramatic improvement in the general symptoms of rheumatoid arthritis They were also able to show that the same results could be obtained by the injection of A C.T H (which again will be dealt with in detail at a later stage) The improvement resulted in some cases in a complete disappearance of general symptoms and was attended by a feeling of well being or euphoria in most of the patients Unfortunately they found that interruption of the treatment was attended by a rapid return of the symptoms and in a few days the patient was in just as bad a condition as before the treatment In some later observations the results appeared in a limited number of patients to be of a little more permanent character

There is no doubt that these classical observations stimulated all the recent interest in the rheumatic diseases

2 **Klemperer Pollack and Baehr (1942) (5) at New York** introduced a generalisation into pathology by combining a number of disease processes as an entity which they called the diffuse collagen disease The underlying pathological process was found to be a fibrinoid degeneration of collagen in this series of diseases which include disseminated lupus erythematosus rheumatoid arthritis periarteritis nodosa, thrombo angitis obliterans and scleroderma—all having the same fundamental pathology An alternative name has been suggested of para rheumatic disease for the above mentioned group

The group of collagen diseases is constantly being added to and there is naturally considerable controversy At the time of writing the following are regarded as being members of this group — serum sickness periarteritis nodosa disseminated lupus erythematosus rheumatic fever rheumatoid arthritis in its various forms scleroderma, dermatomyositis and according to some workers calcinosis Others would like to include Buerger's disease and malignant nephrosclerosis

General Pathology of the Collagen Diseases In the first place the chemistry of collagen itself is incompletely understood It forms the basis of bone and cartilage and the interfibrillary connective tissue It can be broken down to gelatin and various mucopolysaccharides such as chrondroitin sulphuric acid Another constituent is hyaluronic acid which is again a highly complicated substance whose behaviour is controlled by an elaborate system of enzymes of which hyaluronidase or the spreading factor is one From the histological point of view a study of the diseases mentioned above will show that there are in all similar changes in the fibroblasts which are largely composed of collagen They become stained very refractile and the general term for such changes is fibrinoid degeneration The ætiology of this condition is very largely speculative Leaving out for the moment Selye s conception that these diseases are related to stress we can say that various theories have been favoured from time to time Thus bacterial infection has attracted a number of workers from the time of Poynton and Paine in 1900 and 1901 (6 7 8) to the present day work on β hæmolytic group A streptococcus infection

Another suggestion has been that these diseases might be associated with allergy and with immunity Thus Rich and Gregory (1943) (9) claimed to have produced periarteritis nodosa lesions in rabbits by injection of heterologous serum

The role of hormones in the production of this group of diseases will be discussed in relation to Selye s views

In a brief review of the varied collagen diseases we may start with *serum sickness* which made its first appearance after the production and use of diphtheria antitoxin in 1890 The symptoms commence within a week or so of the administration of the serum and consist of enlargement of the lymphatic glands with underlying tenderness and skin lesions such as urticaria itching and macular erythemas may also develop Joint pains may or may not occur The symptoms usually clear up in the course of a few weeks

Periarteritis Nodosa This condition was first described by Kussmaul and Meier in 1866 its clinical diagnosis is extremely difficult and its pathology is essentially a degeneration of the media and intima of the small and medium sized blood vessels Thrombosis hæmorrhage etc can occur again rendering the condition very difficult to diagnose except at post mortem

Disseminated Lupus Erythematosus This is a disease occuring in young women and frequently initiated by exposure to sunlight The skin lesions characteristically have the well known 'butterfly' distribution on the face and may consist of confluent red eruptions, and a swinging pyrexia associated with pains of a rheumatic character in the joints frequently occurs

Scleroderma This condition occurs in the fingers but may involve the whole body There is a characteristic thickening of the skin together with an infiltration of the subcutaneous tissue by collagenous material

Dermatomyositis This is a very rare disease in which it is claimed by many that the underlying pathology is the characteristic collagen degeneration The clinical features consist of pain in the muscles with or without pyrexia Occasionally there may be skin reactions

Calcinosis In this condition calcification of the subcutaneous tissues and connective tissues occurs Its pathology is similar to that of scleroderma but proof that this condition is a collagen disease is not definite

Gout Attempts have been made to include gout in the collagen diseases group, but this appears to be hardly justifiable as any collagen changes that occur are more likely to be due to the primary metabolic defect associated with purine metabolism

3 **Selye at Montreal** has developed a very complex theory to explain the ætiology of the collagen diseases No attempt will be made to give a detailed account of this work as at the present time the views of most workers on this subject are in a state of flux For those requiring a full statement the reader is referred to the massive work entitled Stress by this author (10)

Selye developed the idea that animals react to stress or injury by a certain sequence of physiological reactions which is termed the general adaptation syndrome (or G A S) This consists of a number of phases of which perhaps the most important is the discharge of hormones from the pituitary to act on the suprarenal According to this theory when an animal is subjected to stress or injury the anterior pituitary pours out adreno-corticotropic hormones and the stimulated cortex secretes hormones which Selye divides into two more or less clear cut categories namely those

regulating the electrolyte metabolism which he calls "mineralo-corticoids" and those which influence carbohydrate metabolism and which he calls "gluco-corticoids". According to Selye it is the interplay of the action of these hormones which provides the rheumatoid phenomenon. The experimental evidence on which this rests has been clarified by a number of workers. The fundamental observation was by Selye and Pentz (1943) (11) who showed that unilaterally nephrectomised rats treated with a high sodium chloride intake developed lesions similar to periarteritis nodosa when they are injected with very heavy doses of deoxycorticosterone.

Selye extended this work and showed that giving heavy doses of deoxycorticosterone to animals that had been damaged in various ways such as by adrenalectomy, thyroidectomy, exposure to cold, etc., would cause arthritic changes similar to those in man. On the basis of this Selye put forward the view that the fundamental underlying pathology of the collagen diseases was an upset of the suprarenal cortex produced by the pituitary secretion as a result of stress. This pituitary secretion caused cortical stimulation leading to the outpouring of the mineralo-corticoids which were responsible for the syndrome.

Selye's main work on G.A.S. was prior to the discoveries of Hench and his colleagues and this accounts for the extraordinary confusion of the literature and also for the difficulties of anyone attempting to give an orderly summary. As Marrian puts it in his review of the subject (12) —

"Following the announcement in 1949 by the Mayo Clinic group of the effects of cortisone and of A.C.T.H. in rheumatoid arthritis and rheumatic fever, Selye (1949) (13) re-interpreted his earlier findings concerning the effect of adrenalectomy on the production of experimental arthritis by D.O.C.A. He suggested that it was reasonable to assume that adrenalectomy sensitizes to the toxic effects of D.O.C.A. because it removes the source of endogenous gluco-corticoids and thereby predisposes to the development of a particularly unfavourable gluco-corticoid–mineralo-corticoid balance. In support of this hypothesis he cited a number of earlier observations made by himself and his co-workers suggesting that certain of the effects on living animals of 'mineralo-corticoids', such as deoxycorticosterone are diametrically opposed to those of gluco-corticoids. It may be noted however that this earlier work

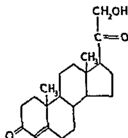
did not include any observations that 'mineralo corticoid' induced rheumatic lesions in animals could be cured by the administration of gluco corticoids In the same communication Selye showed that an experimental 'arthritis could be produced in rats by the injection of formaldehyde into the vicinity of the joints This experimental arthritis he found, could be aggravated by pre treatment of the animals with D O C A and inhibited by cortisone or A C T H"

The whole position of Selye s theory is very difficult to assess today It is for instance very doubtful whether his clear cut distinction between mineralo corticoids and gluco corticoids is justifiable Very careful observations on the pharmacology of compounds such as deoxycorticosterone and so forth tend to show that this clear cut distinction does not hold

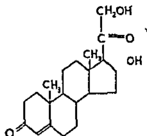
Up to the present it has been possible to isolate and identify those substances capable of prolonging the life of adrenalectomised animals These are as follows — (1) 11 deoxycorticosterone (2) 11 deoxy 17 hydroxycorticosterone (3) Corticosterone (4) 17 hydroxy corticosterone (5) 11 dehydro corticosterone (6) 11 dehydro 17 hydroxycorticosterone (cortisone) In addition to these six substances, adrenal extracts contain the so called 'amorphous fraction the constitution of which is quite unknown and which will also maintain life According to Selye s (1950) views the pharmacology of these compounds can be considered under two headings (*a*) compounds 1 and 2 and the amorphous fraction are concerned with the regulation of electrolyte metabolism and water balance whilst (*b*) compounds 3–6 are concerned almost entirely with the carbohydrate effect Recent investigation has shown that the pharmacological actions of these substances overlap very considerably

Whilst an interpretation of Selye s view must await much more experimental work it is interesting to note that Marrian (1951) (12) and his colleagues have very definitely demonstrated that in established rheumatoid arthritis progesterone metabolism is modified It is known that progesterone is excreted mainly in the form of pregnanediol and when progesterone is administered to a patient with rheumatoid arthritis an abnormally high amount of pregnanediol is excreted The actual interpretation of these results as Marrian points out must be cautious but it would certainly seem

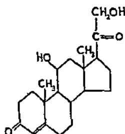
to be interesting evidence that the disease rheumatoid arthritis is associated with some abnormality of steroid metabolism



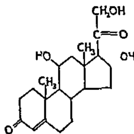
11-deoxycorticosterone



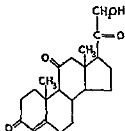
11-deoxy 17 hydroxycorticosterone



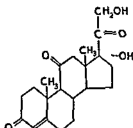
Corticosterone



17 hydroxycorticosterone



11 dehydrocorticosterone



11 dehydro 17 hydroxycortico sterone (cortisone)

Practical considerations Whatever may be the final outcome of the theories which have been discussed above the fact remains that two substances cortisone and A C T H are capable of causing a remission in the symptoms of patients with

collagen diseases particularly rheumatoid arthritis It is proposed to review briefly the chemistry of the two substances

Cortisone This substance is a steroid obtained by Kendall and his colleagues from the suprarenal cortex It is slightly soluble in water and like all steroids soluble in organic solvents and in oil and its production presents problems of immense difficulties Its isolation from the cortex of the suprarenal is a very wasteful process and in any case even were all the glands in the world available the amount of cortisone so produced would be quite inadequate for a fraction of the rheumatoid arthritis population of the world

Unfortunately the methods of synthesis by degradation processes from cholesterol cannot be applied as at the time of writing there is no practical method of introducing either an oxygen or a hydroxy group at the 11 position of the cyclopenteno phenanthrene ring A solution has been found to the production of cortisone by commencing with a bile acid Bile acids have a hydroxy group at the 12 position and by considerable and complex organic procedures this can be transferred to the 11 position

It is no exaggeration to say that the commercial production of cortisone represents one of the greatest triumphs of organic chemistry in its industrial application

A C T H This is extracted from the anterior lobe of pituitaries obtained from the slaughter house It is a protein and its constitution is unknown despite the fact that it has been obtained electrophoretically pure The work of Morris (Cortis Jones Crooke Henly Morris and Morris 1950) (14) has shown that it is possible to ultra filter a low molecular weight component which possesses all the activity of the parent material Similarly Li (1949) (15) has shown that the active extract may be produced by acid hydrolysis or by peptic digestion

A C T H has to be administered intramuscularly or intravenously and it is assumed that it stimulates the suprarenal cortex to produce mineralo corticoids and the gluco corticoids including cortisone

Physiological Action of Cortisone and A C T H in Man

A detailed account of the action of cortisone and A C T H in man is found in a comprehensive article by Sprague (1950) and his colleagues (16) The material used in this investigation was cortisone

acetate administered usually in doses of 100 mg (being equivalent to 89 mg of free cortisone) in the form of a suspension in saline or A C T H obtained in ampoules (containing 25 mg) of which the pressor content and the oxytocin content were known The anti rheumatic effect is not described in this paper except that the authors state that the results of Hench and his colleagues have been confirmed The following are the principal effects of the substances —

Body weight Due to the euphoria and increased appetite produced the body weight was invariably increased in the use of both substances and a marked feature in some cases was a characteristic moon shaped face

Effects on the skin Acne and hairiness were produced in a number of subjects but it is interesting to note that there was no masculini sation of females as is obtained by the use of testosterone in the treatment of inoperable carcinoma Keratosis and the development of cutaneous striæ has been noted and in some instance deep pigmentation occurred

Muscular weakness This developed in a considerable number of patients undergoing prolonged treatment

Changes in electrolyte and metabolism balance A negative balance in nitrogen and potassium was produced by A C T H whilst corti sone caused relatively slight changes It would appear that A C T H produces more of the mineralo corticoid effect than does cortisone Details of individual experiments with these substances will be found in the paper referred to

Effect on menstruation This was very variable in the series of 10 women treated Three became amenorrhœic a fourth missed a period and others remained unaffected In general treatment with both cortisone and A C T H appeared to reduce sexual activity in both male and female patients

Euphoria This again was very variable but occurred very regularly with high dosage consisting of a general feeling of well being with a very definite improvement in mental activity particularly in patients who were seriously ill either with arthritis or some similar condition For example the authors quote the case of a man in the terminal stages of myelogenous leukæmia who when treated with cortisone took a constructive interest in his future despite the fact that he was to die in a very few days time

Blood pressure The general consensus of opinion is that the materials have little effect on blood pressure In a small number of cases there was a significant rise The authors feel however that the substances may induce hypertension is some circumstances

Excretion of urinary steroids Treatment with A C T H causes an increase in the excretion of urinary steroids whereas in the case of cortisone administration the excretion of 17 ketosteroids usually falls unless high doses are given Persistent low levels were observed after very prolonged treatment

Leucocytes Cortisone produced very little consistent change in the circulating leucocytes on prolonged treatment whereas A C T H produces a characteristic disappearance of eosinophils

Effect on cortical function There appears to be evidence that treatment with cortisone produces depression of animal adrenal function It is suggested that this depression is the cause of the lowered excretion of 17 ketosteroids on and after treatment with cortisone

Treatment of Rheumatoid Arthritis with Cortisone and A C T H

The dosage is still mainly unstandardised and arbitrary In the first instance Hench and his colleagues maintained that 100 mg per day in divided doses of 25 mg were necessary to produce an effect though recent workers have found that smaller doses are capable of producing remissions

The actual regimen is not yet established but it is usual to continue the treatment for seven to fourteen days and then to reduce the dose in the hope that the good effects will not pass off With regard to A C T H it is usual to find the effect developing after the administration of the substance for some four to five days and similarly the effect is prolonged for that length of time after the drug has been withdrawn With regard to dosage this again is a matter for experimentation but usually between 40–80 mg are recommended (Prunty 1951) (17) With regard to the type of case treated any member of the collagen group including rheumatic fever is suitable for treatment Contra indications are observed in the following conditions — Myocardial insufficiency diabetes mellitus Cushing s syndrome tuberculosis and advanced renal disease One of the most important contra indications is tuberculosis

for the administration of A C T H and cortisone to patients with tuberculous lesions is frequently followed by dissemination of lesions throughout the body

Treatment of Skin Diseases with A C T H

A very good review of this subject together with an account of a number of cases treated will be found in the paper by Brodthagen Reymann & Schwartz (1951) (18) Pemphigus vegetans was favourably affected and the patient remained clear of symptoms for three months after the treatment had been stopped A case of pemphigus vulgaris was also affected but relapsed almost as soon as the treatment was interrupted In the case of scleroderma improvement was shown in a few cases but some proved to be resistant It is interesting to note that these authors found one patient with periarteritis nodosa resistant despite prolonged treatment The dosage in most of the cases consisted of 75 mg per day for the first week and then a reduction to 25 mg later Frenkel Hellinga and Groen (1951) (19) have described a successful treatment by A C T H of the rare condition known as Sjogren s disease This condition occurs in middle aged women and consists of chronic inflammatory changes in the lachrymal and salivary glands with a diminution in the production of tears and saliva Joint lesions occasionally occur and these are usually of a rheumatoid character The condition relapsed and during treatment the patient appeared to be quite well

Clinical Use of Cortisone

Cortisone acetate is put up in 20 ml vials ready for intramuscular injection each ml containing 25 mg It must be shaken before use and must not be stored in a refrigerator It is injected deeply into the gluteal muscles Before and during treatment of any case certain investigations must be carried out The chest must be X rayed to exclude the presence of tuberculosis An electrocardiogram should be taken with the three standard and nine unipolar leads in case an electrocardiogram is required later to indicate a potassium deficiency Further an abnormal electrocardiogram may return to normal under treatment The patient should be weighed daily The serum sodium and potassium content should be estimated weekly in Addison s disease to check a rise in

the sodium or fall in the potassium level as this is indicative of an overdosage The erythrocyte sedimentation rate should be determined weekly Daily blood pressure readings and tests of the urine for glucose should be made The patient s grip should be tested twice a week by seeing the height to which he can raise the column of mercury in a sphygmomanometer when compressing the rolled up cuff into which a little air has been pumped The joint sensitivity can be tested in cases of rheumatoid arthritis by firm pressure over the affected hands or wrists Grade 0 indicates no pain Grade 1 some pain Grade 2 wincing and Grade 3 the patient pulls his hand away The usual dose for rheumatoid arthritis is 100 mg every eight hours for 3 doses than 100 mg every twelve hours for 2 doses then 100 mg every twenty four hours for seven to fourteen days followed by 100 mg every forty eight hours for fourteen to twenty-eight days It may now be possible to give 50 mg in water by mouth for six days with a weekly intramuscular injection of 100 mg in order to maintain the improvement In Addison s disease smaller doses are usually required such as 12 5 to 25 mg daily intramuscularly

Results We have treated a series of cases including rheumatoid arthritis disseminated lupus erythematosus sarcoidosis Sjogren s disease and Addison s disease The case of Sjogren s disease appeared to do best and maintained the improvement after the cortisone was discontinued The lupus erythematosus and rheumatoid arthritis cases although treated for two to three months quickly relapsed when the treatment was discontinued In sarcoidosis the uveal tract lesions and the salivary gland swellings rapidly improved with the intravenous administration of A C T H in small doses (20) but the X ray shadows in the lungs were not affected

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CHAPTER II

ANTIBIOTIC SUBSTANCES

Penicillin

The last few decades have witnessed the introduction of an increasing number of compounds which whilst showing toxicity of a high order against pathogenic organisms exercise little or no harmful effect upon their human hosts Penicillin was discovered by Fleming in 1929 (1) It is an extracellular product of the metabolism of the mould *Penicillium notatum* and represents one of the first of a series of antibacterial compounds obtained from moulds and bacteria Many of these compounds display potency of a very high order when tested *in vitro* relatively few are sufficiently free of toxic effects on the host to permit their therapeutic application

Antibiotic Activity Penicillin is most active against gram positive and gram negative cocci and gram positive bacilli It is active against spirochætes and possibly some of the larger viruses Sensitivity of various strains of organisms varies widely so that it is important to know the organism responsible for the infection and its sensitivity to penicillin

Administration Penicillin is now available in large quantities and at a fraction of its original cost The yellow amorphous sodium penicillin has been largely superseded by the pure crystalline penicillin G or by pure penicillin G combined with procaine Increasing purification has led to fewer side reactions and there is a general tendency to employ very much larger doses

It is necessary in treating infections with penicillin to maintain an effective bacteriostatic level in the plasma which varies from 0 02–0 20 units per ml This may be achieved by injecting intra muscularly aqueous solutions of penicillin three hourly The rapid elimination of penicillin in the urine prevents this figure being maintained except by very frequent injections

Several attempts have been made to delay the absorption of penicillin from the site of injection This was done by suspending penicillin in sterile white beeswax and arachis oil but the solution

was difficult to suck up into a syringe on account of its viscosity and the beeswax frequently excited undesirable foreign body reactions locally A great advance has been achieved by the introduction of procaine penicillin (2 3) which is an equimolecular compound of penicillin and procaine in a suspending agent It is sometimes combined with a water repellent substance aluminium mono stearate which is said to even out absorption A single dose of procaine penicillin of 300 000 units gives an assayable blood level for twenty four hours Commercially procaine penicillin is often combined with sodium or potassium penicillin G to ensure a satis factory initial blood level

Excretion of penicillin by the kidney can be delayed by some substances which are excreted by the tubules Caronamide (4 5) diminishes renal tubular excretion of penicillin and 3 g by mouth three hourly produces a plasma caronamide level of 15 mg per 100 ml Since the development of preparations such as procaine penicillin there is little general use for caronamide except possibly in treating infections due to resistant bacteria where it is necessary to maintain exceptionally high penicillin blood levels

Penicillin is rapidly destroyed by acid and by organisms such as those found in the gut which produce penicillinase These factors prevent penicillin being administered orally or rectally as a routine Penicillin has been combined with a number of vehicles to neutralise gastric acid and enclosed in capsules to aid absorption from the gut Oral administration is possible and satisfactory blood levels and therapeutic effects have been reported (6 7) but the dose should be about ten times that for intramuscular injection It may be used in children and infants or where the organism is highly sensitive to penicillin

Toxic Effects Penicillin has maintained its promise of being almost non toxic in therapeutic doses Skin reactions especially urticaria are the commonest effects although drug fever and swelling of the joints may occur The reactions usually come on after treatment has been maintained for some time but some patients may develop allergic reactions very rapidly When penicillin is given by mouth glossitis and stomatitis may occur and there is often secondary invasion with saprophytic fungi These reactions may be reduced by giving the Vitamin B complex to patients who are taking penicillin by mouth When penicillin is

used topically for skin diseases a sensitisation dermatitis may occur and it is important to discontinue penicillin should this happen

There is usually no indication to discontinue treatment on account of the milder allergic effects and they may be controlled by the administration of one of the anti histamine groups of drugs For the rare severe general reactions desensitisation may be attempted by giving very small doses of penicillin subcutaneously such as 400 units three times a week and increasing the dose gradually until a full therapeutic dose can be injected without any side effects (8)

Preparations of Penicillin Many of the earlier preparations of penicillin have been replaced by the more recently developed ones Penicillin cream and solutions of penicillin should be kept in a refrigerator Aqueous solutions of procaine penicillin retain their potency for seven days at room temperature and twenty one days in a refrigerator The other preparations of penicillin keep for up to two years in the dry state in cool dry conditions The date of expiry is usually printed on the vial

Powder Sodium or potassium penicillin G is put up in rubber capped vials containing 100 000 200 000 500 000 and 1 000 000 international units Calcium penicillin which is used for surface applications is put up in containers of 1 10 or 50 Mega units (1 Mega unit = 1 000 000 units)

Procaine Penicillin This is put up as — Penicillin G procaine salt (300 000 units per ml) Penicillin G procaine salt in aqueous suspension (300 000 units per ml) Penicillin G procaine salt + penicillin G (300 000 units of procaine penicillin G per ml + 100 000 units of penicillin G)

Tablets for Oral Use 10 000 20 000 50 000 and 1,000 000 units of penicillin G per tablet

Lozenge 500 units calcium penicillin per lozenge

Chewing Gum 5 000 units calcium penicillin

Creams 500 mg calcium penicillin/g Lanette wax base with 1 1% phenoxetol

Ointments Calcium penicillin 50 000 units and ointments of alcohols 100 g They contain 500 units penicillin/g

Powder for Local Application 5 000 units/g of calcium penicillin combined with a sulphonamide or starch or talc base

Eye Drops 500–2 500 units per ml

Eye 'Tabloids' 250 units per tabloid
Eye Ointment 25 000 units penicillin G per g

Clinical Applications

Intramuscular Injection A dose of 50 000–250 000 units of penicillin is given every three hours

The dose is prepared by adding 1–2 ml of sterile pyrogen free water to the powder The vial should then be gently rotated vigorous shaking is unnecessary and may cause frothing The syringe and needle should be boiled in plain water and kept in distilled water They must never be placed in antiseptic or spirit as these substances destroy penicillin The skin round the site for intramuscular injection may be prepared with soap water ether or Cetrimide For ordinary intramuscular injection an Arnold No 6 serum needle may be used

Procaine penicillin is administered by intramuscular injection only The dose is 300 000–600 000 units of procaine penicillin every twelve hours A 5 ml syringe fitted with a No 20 S W C needle is suitable and 2–4 ml of sterile distilled water are injected through the rubber cap at the base of the vial This is unnecessary with the aqueous suspension of procaine penicillin The vial is shaken vigorously to suspend the particles It is advantageous to build up a positive pressure by injecting a syringe full of air into the vial The required dose (1–2 ml) is withdrawn and injected into the patient

Powder for Local Treatment Dry penicillin powder made up with sterile sulphathiazole is used for wounds and abrasions The powder is blown on to the surface with an insufflator to form a thick hoar frost The application should be repeated every twenty four hours but must not be continued for longer than five days owing to the risk of the development of skin reactions due to sensitivity to the sulphonamide This method is of most use in relatively dry surfaces as blood or exudate tends to wash away the powder

Ointments and Creams These are used for local application in certain skin or eye infections

Lozenges or Pastilles These are useful in treating infections of the mouth and throat One pastille should be sucked every hour for 8 hours

Eye Drops In certain eye infections drops containing 500-2 500 units per ml every one or two hours are beneficial

Inhalation Penicillin may be given by inhalation in the treatment of bronchitis and bronchiectasis by means of an inhaler such as the Collison 20 000 to 100 000 units in 2 ml of distilled water are put into the bulb for each inhalation and the 'mist' inhaled every two hours daily through the oro nasal mask furnished with an expiratory valve The vaporisation is effected by oxygen and takes about fifteen minutes The rubber tubing of the apparatus should not be a synthetic preparation which inactivates the penicillin

Solutions These can be made up with distilled water or saline at 1 000 units per ml for instilling twelve hourly through fine rubber tubes into wounds which have been closed for injecting into abscess cavities after aspiration and as a spray for skin and mucous membrane infections Solutions of a strength of 1 000 to 2 000 units per ml are suitable for injecting into the cerebro spinal pleural pericardial and joint cavities and 10 000 units per ml for intratracheal injection

Septicæmia Successful treatment of septicæmia whether of staphylococcal or streptococcal origin necessitates not only the complete sterilisation of the blood but also the elimination of primary and secondary foci of infection It is recommended that 100 000–250 000 units of penicillin G should be injected intra muscularly every three hours

Sub Acute Bacterial Endocarditis Patients should be given at least 2 000 000 units of penicillin G a day for six weeks by inter mittent intramuscular injection every three hours If there is a relapse or an increase in resistance of the organism 5 000 000 units a day for eight weeks or more should be given (9)

Meningitis Successful treatment of meningitis by the ad ministration of penicillin and sulphonamides has been reported by many workers including Rosenberg and Arling (10) and Daniels et al (11) Staphylococcal infections of the meninges are usually secondary to mastoid and sinus foci Immediate surgical drainage of such centres followed by local treatment with penicillin solution is of considerable value in reducing the chance of reinfection Smith Duthie and Cairns (12) advise the treatment of uncom plicated pneumococcal meningitis as follows After finding pneu

mococci in the cerebro-spinal fluid 8 000 to 16 000 units of penicillin are injected intrathecally with smaller doses for infants The penicillin is in pyrogen free saline solution 2 000 units per ml Fifteen thousand units of penicillin are injected intramuscularly every three hours to control any primary infection or septicæmia Sulphadiazine is also given by mouth or nasal tube first 4 g and then 2 g every four hours an adequate fluid intake being ensured to prevent renal block. After twelve hours a second intrathecal injection of penicillin is given If the fluid is now thicker a third puncture should be made twelve hours later to detect any incipient ventricular block which might require intraventricular injections of penicillin Daily injections of penicillin into the theca must be continued for five days the intramuscular injections are discontinued after five days and the sulphadiazine reduced to 1 g four hourly after five days and maintained at this dose for a week

If the cerebro spinal pathway becomes blocked by pus in sites such as the aqueduct penicillin should be injected into the lateral ventricles through parietal burr holes After draining the pus the penicillin can be injected through an indwelling catheter In all cases in which intrathecal injections are required non irritating preparations of penicillin should be used and great care taken in the aseptic technique In meningococcal and streptococcal meningitis similar amounts of penicillin are injected intrathecally every twenty four hours until the cerebro spinal fluid is free from organisms and intramuscular injections of 100 000 units are given every three hours for seven days

Pneumonia In the treatment of pneumonia there is little to choose between penicillin and the sulphonamides except in cases of sulphonamide resistance especially those due to staphylococci or where the lower toxicity of penicillin is advantageous The usual dose for an adult is 50 000–100 000 units every three hours for seven days for a child 5 000 units per year of age Primary atypical (virus) pneumonia does not respond to penicillin

Empyema In the treatment of empyemata due to pneumococci streptococci or staphylococci but not in cases of Bact coli infections also in infected hæmothorax following injury intra pleural instillation is the method of choice Penicillin solution containing 1 000 units per ml should be injected into the pleural space following aspiration of pleural fluid The amount of penicillin injected

depends on the size of the empyema cavity Cavities holding 10 to 20 oz of fluid should receive 60 000 units of penicillin total empyemata 120,000 units and 30 000 units for small pockets Three to four aspirations and instillations generally suffice Thoracotomy is usually required to evacuate blood clot fibrin and pus Local penicillin treatment after rib resection is often unsatisfactory and penicillin is then best given by eight hourly intramuscular injections of 100 000 units

Bronchiectasis The cavities are emptied as far as possible by postural drainage followed by the intratracheal injection of penicillin The skin and subcutaneous tissues are anæsthetised in the neck between the cricoid cartilage and the first ring of the trachea with 2 ml of 2% procaine Two ml of 2% Amethocaine are then injected rapidly through a No 14 needle into the trachea and the patient told to avoid coughing The syringe is detached from the needle the patient placed in the optimum position for fluid to run down the trachea to the affected part of the lung and then 8 ml of penicillin solution containing 10 000 units per ml are rapidly injected through the needle which has been left *in situ* in the trachea This treatment repeated daily for seven days has resulted in considerable clinical improvement Alternatively the penicillin can be given every two hours by inhalation

Chronic Bronchitis In some cases the amount of sputum can be very considerably reduced and the breathing made easier by the inhalation of penicillin as described above

Infections of the Skeletal System Osteomyelitis and coccal infections following compound fractures respond well to both penicillin and sulphonamide therapy It is on record that cases of chronic osteomyelitis of some years duration have been cleared in a relatively short period by penicillin treatment Sterile blood cultures may be obtained following intravenous supported by intramuscular injection but the intramedullary route is obviously recommended for resistant strains Grace and Bryson (13) treated *7 cases of chronic osteomyelitis with 1 failure The penicillin was* given intramuscularly in doses of 30 000 units every three hours for ten to twenty-one days and simultaneously 4 000 to 20 000 units of penicillin per ml were applied locally into the sinus tract In open wounds aspiration of exudate and local administration of penicillin and sulphonamide powder accelerate the recovery Kirby

and Hepp (14) have emphasised the risk of secondary meningococcal infection from osteomyelitis of the facial bones and recommend penicillin rather than sulphonamides in order to reduce the risk of reinfection Penicillin treatment of osteomyelitis should be followed by surgical removal of infected or necrotic bone Infections by streptococcal and staphylococcal organisms located in bone joints and synovial membranes respond well to prolonged penicillin treatment In contrast chronic rheumatoid arthritis even when associated with recognised coccal strains known to be penicillin sensitive fails to respond to what must be regarded as massive doses of the drug Boland Headley and Hench (15) who have given careful attention to classical cases of arthritis report complete failure

Florey and Florey (16) recommend the daily application of penicillin in the wound following mastoidectomy

Gonorrhœa in the Male The efficacy of penicillin treatment in gonorrhœa has been established beyond doubt Even when examined over a period of several weeks patients show no recurrence of symptoms Welch et al (17) report the superiority of penicillin X in the treatment of gonorrhœa by single intramuscular injections of 25 000 units but many other workers suggest that 100 000 units of penicillin or even more are essential to ensure a permanent cure in all cases Most strains of the organism are sensitive to sulphathiazole treatment but are prone to, develop sulphonamide resistance Fortunately the sulphonamide resistant strains which are encountered show no penicillin resistance Five injections of 20 000 units every two hours are usually sufficient to effect a cure The difficulty of maintaining a sufficiently high blood concentration by one injection in out patients who cannot attend for the five injections has been overcome by Romansky and Rittman (18) who recommend a beeswax peanut oil mixture as the medium for injection By this means they cured 11 out of 12 patients suffering from gonococcal urethritis with a single injection of penicillin of 40 000 to 66 000 units Romansky et al (19) obtained 100 / of cures in 75 cases of gonorrhœa in males by a single oil injection of 175 000 units of penicillin Whatever treatment is given the patient should be re examined for infection after two and four months A single dose of 600 000 units of procaine penicillin is usually effective

Gonorrhœa in the Female This responds equally well to

penicillin therapy, symptoms disappearing in the majority of cases following the administration of 100 000 units

Syphilitic Infections In primary and secondary syphilis penicillin therapy has proved equally satisfactory The results obtained by many workers compare well with those using massive arsenical therapy, without exposing the patient to the toxic effects and idiosyncrasies of the latter drugs Whether penicillin is given intramuscularly or intravenously a continuously high concentration in the blood must be maintained Mahoney et al (20) in an early communication showed the disappearance of the spirochætes and clearing of serological reactions following the administration of 25 000 units every four hours for eight days Moore et al (21) and Norcross (22) confirm this finding and have reported apparent recovery from primary and secondary syphilis following treatment with much lower quantities of penicillin Leifer (23) from his experience in treating 96 cases concluded that early syphilis can be cured by intramuscular injections of 20 000 units every three hours for seven and a half days giving a total of 1 2 million units Lourie et al (24) have reported favourably on a method for the ambulatory treatment of early syphilis Three intramuscular injections of 600 000 units were given at hourly intervals on five consecutive days The penicillin was dissolved in 5 ml water The treatment of cardio vascular syphilis with penicillin has proved unsatisfactory in the hands of Dolkart and Schwemlein (25) the injections having to be abandoned owing to cardiac pain The simultaneous administration of arsenicals and penicillin has been claimed to have a synergistic effect and to be superior to straight forward penicillin therapy The Committee on Medical Research and the United States Public Health Service (26) reported in 1946 on the treatment of early syphilis with penicillin They found that the percentage failure at the end of eleven months after treatment varied from 15% when 2 400 000 units of penicillin had been given to 62% with 600 000 units Patients treated during the first week of disease did twice as well as those treated after two months or later Simultaneous treatment with arsenic or bismuth increased the number of patients cured when total amounts of penicillin were given varying between 300 000 to 1 2 million units Crystalline penicillin G is superior to amorphous penicillin in both experimental animals and man McElligott (27) recommends for primary and

early secondary syphilis a course of 8 daily injections of procaine penicillin followed by 10 weekly injections of bismuth suspension, each injection being equivalent to 0 2 g of bismuth metal

Gas Gangrene The administration of penicillin to subjects with wounds infected by organisms producing gas gangrene has been reported from several sources No clear indication of the effectiveness of penicillin alone is possible since the administration has always been accompanied by injections of antitoxin local administration of sulphonamides and where necessary by surgical operation Although Cutler (28) has reported 4 cases of gas gangrene infections following compound fractures which failed to respond to intravenous injection of penicillin the consensus of opinion in this matter is that local administration of penicillin is at least as effective as any preceding method and that intravenous administration of the drug prior to operation is an effective means of localising the area of attack

Tetanus *In vitro* Cl tetani is sensitive to penicillin Altemeier (29) did not find that penicillin had any beneficial influence on the course of established clinical tetanus This is to be expected as penicillin has no known effect on the tetanus toxin

Agranulocytosis Boland et al (30) have shown that penicillin *is of value in treating agranulocytosis due to various causes as* death generally results from secondary infections rather than from the suppression of bone marrow function If the infection can be checked the bone marrow is given a chance to recover The agranulocytosis may be due to such causes as sulphonamides mapharsen gold salts thiouracil etc

Actinomycosis Hamilton and Kirkpatrick (31) successfully treated 2 cases of cervical actinomycosis with three hourly intramuscular injections of penicillin the dose varying from 25 000 to 33 000 units and a total of between 5 and 6 million units being given We treated a case of actinomycosis of the pleura in which the organism was penicillin sensitive *in vitro* with intramuscular and intrapleural injections of penicillin but the disease proved fatal in a few weeks

Wounds and Burns The low toxicity of penicillin together with its high bacteriostatic action makes it pre eminently suitable for local administration at the wound sites especially when combined with a sulphonamide

Skin Diseases Penicillin may be used as a cream or a spray containing 500 units of penicillin per ml It is of value in skin infections caused by staphylococci and streptococci Impetigo and sycosis barbæ respond well to local treatment but if cases relapse a course of intramuscular injections is useful in reaching the deeper parts of the hair follicles Eczema with secondary infection is improved by penicillin locally When the infection has cleared up it is important to treat the underlying eczema Blepharitis responds well to penicillin cream The spray or cream should be applied three or four times daily The cream must be smeared on with a sterile spatula or the handle of a spoon which has been boiled Carbuncles cellulitis erysipelas and cavernous sinus thrombosis are best treated by intramuscular injection of 20 000 units every three hours for five days Good results have followed the use of penicillin cream in pemphigus neonatorum but intramuscular injections are needed in severe cases

Eye Infections Superficial eye infections caused by penicillin sensitive organisms can be treated by instillation every one to two hours of drops containing 500–2 500 units of penicillin per ml by the application of penicillin ointment or by ophthalmic Tabloids containing 250 units Blepharitis conjunctivitis ophthalmia neonatorum infective keratitis and corneal ulcers respond well Penicillin is especially of value in the treatment of sulphonamide resistant staphylococcal infections

Throat Infections For streptococcal tonsillitis penicillin lozenges may be sucked every hour for 8 doses or the throat sprayed two hourly with a solution containing 1 000 units per ml In Vincent s infections it is usually necessary to give a course of intramuscular injections

AUREOMYCIN

Aureomycin was discovered in 1948 by Duggar (32) It is produced from a fungus *Streptomyces aureofaciens* which forms a golden yellow pigment during its growth The chemical formula of aureomycin has not yet been determined nor has it been synthesised its molecular weight is known to be about 500

Pharmacology Aureomycin is a yellow crystalline substance which is soluble in acid or alkaline solution but is sparingly soluble in water A 2% aqueous solution of aureomycin hydrochloride has

a pH of 4 5 In the dry state the drug is stable but in solution it rapidly loses its activity especially in the presence of serum or blood When given by mouth or per rectum aureomycin is rapidly absorbed into the blood and passes into the cerebro-spinal peritoneal and pleural fluids It is found in bile urine and milk and probably crosses the placental barrier (33) Doses of 5–10 mg per kg body weight of aureomycin given orally at six hourly intervals maintain measurable blood levels It appears rapidly in the urine in high concentration and has been detected fifty five hours after a single dose of 0 5 g

Dosage and Administration Aureomycin is supplied in capsules containing 25 50 or 100 mg and in vials for intravenous use containing 100 or 500 mg For adults give either 25 mg per kg body weight every twenty four hours in divided doses or 500 mg every six hours This dosage should be maintained until there is a satisfactory clinical response and it should be continued for three to five days afterwards If there is no improvement within forty eight hours of starting treatment with aureomycin the dose should be increased to 500 mg every three hours and reduced to 500 mg every six hours when the patient becomes afebrile The dosage for children is 80 mg per kg body weight every twenty four hours in divided doses every six hours

Aureomycin may be injected intravenously as an emergency measure in a severely ill patient but this route should be discontinued when the patient can take the drug orally as there is a risk of thrombophlebitis at the site of injection For intravenous use 500 mg of aureomycin may be injected twice daily This dose is dissolved in 50 ml of physiological saline and should be prepared immediately before use The drug must be injected slowly In eye infections aureomycin may be applied locally as a 0 5 % solution of aureomycin borate 1–2 drops being instilled into each conjunctival sac every two hours

Toxic Effects Aureomycin is relatively non toxic and side reactions have decreased with the production of purer preparations It may cause nausea vomiting diarrhoea and flatulence There is seldom any indication for discontinuing the drug and the side effects may be reduced by giving half the six hourly dose every three hours *As with chloramphenicol patients receiving aureo*mycin may develop mucous membrane and dermal lesions of the

type seen in deficiency of the Vitamin B complex Women may develop vaginitis and vulvitis which responds well to synthetic oestrogens by mouth Soreness of the mouth and tongue and perianal exudation and fissuring (which is probably due to secondary invasion with *Candida albicans*) can be prevented or minimised by giving adequate doses of the B complex (34)

Antibiotic Activity Aureomycin has been shown to act bacteriostatically and at higher concentrations as a bacteriocide *in vitro* and *in vivo* against a wide variety of pathogens In general it is effective against gram negative and gram positive cocci, gram negative and gram positive bacilli, the rickettsiæ and some viruses

Organisms do not normally become resistant to aureomycin except the Proteus vulgaris which has been shown to do so readily

Clinical Applications

Staphylococcal Infections During the past few years staphylococci have been encountered which are resistant, or relatively so to penicillin Such strains of staphylococci are found especially in patients with infections developing from ward cross infection and in patients who have had penicillin therapy These penicillin resistant staphylococci can cause epidemics in hospital communities Penicillin resistant staphylococci are susceptible to aureomycin (35) Some strains of staphylococci have shown an increase in resistance to aureomycin (36 37 38) and aureomycin resistant staphylococci may be a problem of the future Staphylococcal infections in general whether treated by penicillin or aureomycin should be treated by full courses of the antibiotic in maximum dosage Staphylococcal septicæmia meningitis endocarditis pneumonia osteomyelitis pyodermia mastitis and other miscellaneous infections caused by staphylococci or where staphylococci were playing a part in the disease have been shown to respond rapidly and effectively to aureomycin therapy

Streptococcal Infections Aureomycin is effective in infections caused by β hæmolytic streptococci Streptococcal pneumonia erysipelas cellulitis scarlet fever tonsillitis endocarditis meningitis and septicæmia respond satisfactorily to aureomycin Aureomycin is very valuable in treating infections caused by Str fæcalis and Str viridans

Pneumococcal Infections Pneumococcal infections involving lungs and meninges in children and adults respond satisfactorily to aureomycin in standard dosage

Bacterial Pneumonias Aureomycin is of great value in the treatment of bacterial pneumonia Infections where the pre dominant organism is a pneumococcus staphylococcus strepto coccus H influenzæ or K pneumoniæ (Bact friedlanderi) respond well It is especially valuable in treating pneumonia due to penicillin resistant staphylococci and in mixed viral and bacterial pneumonias Herrell (39) states that since aureomycin produces good results in bacterial and non bacterial pneumonia it is the treatment of choice for all types of pneumonia Aureomycin is given in doses of 500 mg six hourly by mouth There is rapid symptomatic improvement in twelve to forty eight hours and this is followed in three to five days by radiological resolution

Primary Atypical (Non Bacterial) Pneumonia Several authors have reported the favourable effect of aureomycin in primary atypical pneumonia (40 41 42 43) The temperature falls in forty-eight hours and there is symptomatic improvement with radiological resolution in 3–7 days The effect of aureomycin has been dramatic in cases which were alarmingly ill with high fever cyanosis dyspnœa and delirium Penicillin and sulphadiazine had not improved these patients

Psittacosis Experimental and clinical reports of treatment of psittacosis have been favourable

Rickettsial Infections Epidemic and murine typhus scrub typhus Rocky Mountain Valley fever (44 45 46) and Q fever have all been shown to respond very effectively to aureomycin (47)

Urinary Tract Infections In the absence of impaired renal function aureomycin is excreted in the urine in high concentration two to eight hours following oral administration It is effective in infections of the urinary tract due to Bact coli Bact aerogenes K pneumoniæ (Bact friedlanderi) Str fæcalis and Pseudomonas pyocyanea It has some action against Proteus vulgaris but this organism may become highly resistant during treatment A com bination of streptomycin and aureomycin is valuable in treating mixed infections 500 mg of aureomycin are given every six hours until the infection is controlled and this dose should be continued for several days to prevent recurrence Acute infections respond

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well Chronic infections tend to recur unless mechanical obstruction to the outflow of urine and calculi is dealt with surgically Aureomycin has been shown to be effective when other antibiotics and urinary antiseptics have failed and it is especially valuable when strains of bacteria have developed resistance to other antibiotic agents (48 49 50 51)

Brucellosis Many clinical reports have indicated the value of aureomycin in the treatment of brucella infections Acute infections with Br abortus Br melitensis or Br suis respond promptly to 250 mg every three hours for twelve to fourteen days If there is a relapse a further course may be given but 80 % of 110 cases of chronic brucellosis treated by Harris resulted in complete recovery (52) Debono (53) has drawn attention to an initial rise in temperature lasting one to two days which is followed by a fall in pyrexia and clinical improvement Herrell and Barber (54) and Rosenbaum and Reveno (55) believe that aureomycin combined with dihydrostreptomycin is the treatment of choice in brucella infections During the treatment of brucellosis with aureomycin where rather larger doses are needed than for other infections Vitamin B complex should be given by mouth as this reduces the incidence of mucous membrane and dermal side effects which may develop

Salmonellæ and Shigellæ Infections Aureomycin has not been shown to have an important place in the treatment of these infections Most reports have been equivocal although isolated good reports have appeared

Whooping Cough Bradford and Day (56) found that aureomycin had a bacteriostatic effect on Hæmophilus pertussis and that daily doses of 10–100 mg per kg body weight protected mice given intranasal instillations of 250 M L D of the organism Clinical reports (57 58) have shown that aureomycin brings about symptomatic improvement with reduction in frequency and intensity of paroxysms vomiting and cyanosis

Skin Infections Aureomycin has been reported of value in treating herpes simplex Kaposi s varicelliform eruption eczema vaccinatum dermatitis herpetiformis molluscum contagiosum pemphigus and herpes zoster Aureomycin is given by mouth in usual dosage and this may be combined with aureomycin locally

Liver Disease In a controlled series aureomycin did not appear to have any effect in acute viral hepatitis (59) but some

authors consider it of value in treating chronic hepatic disease following viral hepatitis and in hepatitic coma from various causes (60 61) Aureomycin prevented the development of cirrhosis in rats which had been fed on a diet (*i e* high fat low protein) which produces cirrhosis experimentally (62)

Acute Amœbiasis Aureomycin has been shown to be of value in acute intestinal amœbiasis (63) but more work is needed to assess the place of aureomycin in the treatment of amœbiasis

Gonorrhœa Chen et al (64) reported cure of acute gonorrhœa in males and females in 98% of cases 3–6 g was given daily for three days

Syphilis Clinical work on aureomycin in syphilis is still in the experimental stage It appears to produce a satisfactory initial decline in serological titre and sterilises surface lesions in about forty hours (65) Willcox (66) has obtained gratifying clinical results and recommends further trials of aureomycin in syphilis

Lymphogranuloma Venereum Good results have been obtained in a number of cases of this infection treated with aureomycin There is retrogression of buboes and marked clinical improvement after four days therapy Runyan et al were unable to demonstrate the virus in the healing buboes after three days treatment (67)

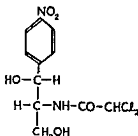
Chancroid Willcox reports aureomycin to be of value in the treatment of chancroid (68)

Other Infections Aureomycin has been reported to be of value in the treatment of the following diseases — Rat bite fever lepto spirosis fusospirochetosis trichomonas vaginalis ulcerative colitis mumps infective mononucleosis lymphocytic choriomeningitis actinomycosis epidemic keratoconjunctivitis dendritic keratitis conjunctivitis and peritonitis

CHLORAMPHENICOL

Chloromycetin (as first called) was produced from cultures of *Streptomyces venezuela* in 1947 by Burkholder and his colleagues at Yale University (69) It was subsequently synthesised by Crooks and his co workers at the Parke Davis Laboratories and the sub stance is now known as Chloramphenicol and is the first antibiotic to have been prepared synthetically

Chemical Composition Chloramphenicol is D (–) *threo* 1 *p* nitrophenyl 2 dichloracetamido propane 1 3 diol



Pharmacology Chloramphenicol is a white crystalline powder which is sparingly soluble in water It is stable in aqueous solution at room temperature for twenty four hours It is rapidly absorbed from the gastro intestinal tract An oral dose of 8 g of chloram phenicol produces a maximum blood level in two hours after eight hours this level is approximately halved and the drug is not detect able after 24 hours (70) Chloramphenicol is absorbed per rectum and may be administered by this route in infants or patients who cannot swallow The capsule is pricked at both ends before being introduced into the rectum

Chloramphenicol is found in bile cerebro spinal fluid and body fluids and it crosses the placental barrier (71) It is principally excreted in the urine

Dosage Chloramphenicol is supplied in capsules each con taining 0 25 g For adults the average dose is 50 mg per kg of body weight daily (in divided doses) The interval between doses should not exceed eight hours to ensure the maintenance of effective therapeutic levels in the blood When the patient has become afebrile and there is a satisfactory clinical response the dose may be halved *i e* 25 mg per kg body weight per day This dose should be continued for two to seven days after the infection has been controlled clinically In infants and children 100 mg per kg body weight should be given at four to six hourly intervals followed by 50 mg per kg body weight for two to seven days

Toxic Effects Chloramphenicol is generally well tolerated The drug has a bitter taste and may cause anorexia nausea vomit ing diarrhœa sore tongue and muscle fatigue Transient ophthal moplegia has been reported in children No serious or permanent toxic effects have occurred and the gastro intestinal upset clears in a day or two when the drug is discontinued In children the

bitter taste may be masked by mixing the drug with honey jam or blackcurrant purée Chloramphenicol inhibits the growth of the normal intestinal flora which may be responsible for the bio-synthesis of the B_2 group of vitamins therefore it is advisable to give the patient a preparation containing all the B_2 complex The normal bacterial flora of the intestinal and respiratory tract inhibit the growth of yeasts so children suffering from thrush should not be given chloramphenicol Chloramphenicol may be given with sulphonamides penicillin streptomycin and aureomycin without incompatibility and it is effective against organisms within its range which have become resistant to other antibiotics

Antibiotic Activity Chloramphenicol is effective against gram positive and gram negative pathogens the rickettsiæ and certain of the larger viruses

Clinical Applications

Typhoid Fever Chloramphenicol has been shown to have a rapid curative action in this infection Woodward et al (72) in a controlled series of patients suffering from typhoid fever showed the effectiveness of chloramphenicol Defervescence of fever occurs in two to three days the urine and stool cultures become negative for S typhi and the titre of the Widal reaction falls within ten to fourteen days Perforation of the bowel or bleeding from a typhoid intestinal ulcer may occur although the infection is well controlled clinically Convalescence therefore should be careful and prolonged If stool or urine cultures become positive for S typhi during this period or if there is a clinical relapse a further course of chloram phenicol should be given The use of chloramphenicol has been disappointing in chronic carrier states with or without gall bladder involvement Negative stool cultures are produced temporarily but relapse invariably occurs

For an average adult patient an initial loading dose of 3–4 g is given over a period of one hour This is followed by 3–4 g daily in divided doses at eight hourly intervals until the patient becomes afebrile then 1 5 g daily eight hourly for fourteen days Bacterio logical or clinical relapse can be treated by a further course (3 g daily at eight hourly intervals for 3–5 days)

Paratyphoid Fever Paratyphoid fever responds to chloram phenicol (73) in a manner similar to typhoid fever save that larger

doses i e 100 mg per kg body weight daily in adults are recommended Results with chronic carriers have been disappointing

Salmonellæ Infections This group of organisms is extremely sensitive to chloramphenicol and it has been found effective in gastro enteritis due to S oranienburg S newport S typhi murium S choleræ suis and S san diego in children and adults 50–75 mg per kg body weight per day eight hourly for three to five days produces clinical and bacteriological cure

Shigellæ Infections This group of organisms is responsible for *bacillary dysentery and although results of treatment with* chloramphenicol have been encouraging the more recently developed members of the sulphonamide group of drugs remain the first choice

Rickettsioses Chloramphenicol represents an important advance in the treatment of the Rickettsioses which include epidemic and endemic or murine typhus fever scrub typhus rickettsial pox Rocky Mountain spotted fever and Q fever

Smadel et al (74) treated cases of epidemic typhus murine typhus and scrub typhus successfully with doses of chloramphenicol of 50–60 mg per kg body weight followed by 0 2–0 3 g two to four hourly for twelve days Q fever has been reported in England recently (75) and this is the only one of the Rickettsioses likely to be found in the British Isles A case of Q fever in a pathologist was successfully treated by Whittick (76) who gave an initial dose of 3 5 g of chloramphenicol followed by 0 25 g every three hours for thirty six hours

Primary Atypical Pneumonia This infection characterised by *fever malaise myalgia headache pleuritic pain cough* and often blood stained sputum with patchy consolidation in the lungs has been successfully treated by chloramphenicol There is rapid symptomatic improvement in one to two days which is followed by radiological resolution 50–75 mg per kg body weight should be given daily in divided doses at eight hourly intervals for three to five days (77 78 79)

Whooping Cough Clinical observations have shown that chloramphenicol reduces the cough plate count controls the pyrexia and toxæmia and converts an intractable and lengthy disease into a mild ailment Gray (80) observed that chloramphenicol limited the course of the infection irrespective of the stage at which treatment is started The initial dose is 50–100 mg per kg body weight

followed by maintenance doses of 0 125–0 3 g six hourly for seven days (81 82 83)

Brucellosis Acute brucellosis due to Br abortus Br melitensis and Br suis respond well to chloramphenicol in doses of 50–100 mg per kg body weight followed by 50 mg per kg body weight six hourly until the fever subsides then 0 25 g three hourly for five days If there is a relapse this course should be repeated Chronic brucellosis responds satisfactorily initially but careful follow up studies are necessary before complete cure can be proved (84 85 86)

Bacterial Pneumonia Lobar pneumonia and bronchopneumonia due to Str pneumoniæ K pneumoniæ (Bact friedlanderi) Staph pyogenes hæmolytic streptococci and H influenzæ respond to chloramphenicol especially in infants and children The patient becomes afebrile in forty-eight hours and radiographic findings usually resolve in a week Bronchial obstruction sterile effusion and empyemata should be treated by the appropriate methods For adults dosage is 3 g daily for five to eight days For children 100 mg per kg body weight should be given at four to six hourly intervals followed by 20–30 mg per kg body weight for seven days or more when the fever has subsided (87 88)

Urinary Infections A high concentration of chloramphenicol is found in the urine after the usual doses by mouth and it has been found of value in treating chronic and acute infections of the urinary tract due to the coli aerogenes group of bacteria K. pneumoniæ (Bact friedlanderi) and many other strains of enterococci and streptococci Proteus vulgaris and Pseudomonas infections may not respond In mixed infections due to gram positive cocci and gram negative bacilli chloramphenicol may be combined with penicillin streptomycin or sulphonamides Recurrence of infection is likely to occur unless mechanical obstruction and anatomical defects are dealt with surgically The doses recommended are an initial daily dose of 2–3 g divided into 2 or 4 doses followed by 0 25 g 4 times a day for five to seven days after the urine has become sterile (89)

Gonococcal Infections Acute gonorrhœa in males and females is satisfactorily treated by 3 g of chloramphenicol initially followed by 1 g eight hourly for two to three days

Syphilis Chloramphenicol has a weak action against Treponema pallidum *in vitro* but it has been found effective in treating acute primary and secondary syphilitic lesions It is not yet possible to

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$C_{21}H_{37-39}N_7O_{12}$ It consists of a basic substance streptidine ($C_8H_{18}O_4N_6$) which is joined to a nitrogen containing disaccharide called streptobisamine $C_{13}H_{21-23}O_9N$ by a glucoside linkage

Streptomycin is readily soluble in water but it is poorly soluble in organic solvents It is stable in the dry state at room temperature for a year and aqueous solutions keep for two months at a temperature of 38° C

Streptomycin is poorly absorbed from the intestinal tract even in large doses and most of the drug can be recovered from the fæces It can only be used by mouth therefore in the treatment of intestinal infections

The method of choice for administration is by intermittent intramuscular injection A dose of 1 g gives a maximum blood level of 20–50 μg per ml falling to 4–10 μg per ml and less than 1 μg per ml at twenty four hours When streptomycin is given intramuscularly it diffuses into pleural and peritoneal fluids but poorly into the cerebro spinal fluid except when there is inflammation of the meninges Streptomycin is mainly excreted unchanged in the urine most of the dose being excreted within twenty four hours

Dosage Streptomycin is supplied in the dry state in rubber capped vials each containing 1 g of streptomycin hydrochloride streptomycin sulphate or streptomycin calcium chloride complex The powder is dissolved by injecting 5 ml of pyrogen free distilled water into the vial with a syringe and needle When the streptomycin has completely dissolved it is sucked up again into the syringe and injected into the upper and outer quadrant of the buttock the upper third of the lateral aspect of the thigh or into the deltoid muscle For adult infections (excluding tuberculosis) 1 g should be injected intramuscularly every six hours This dose should be continued for three to four days or until there is a satisfactory clinical response when the dose may be reduced to 0 5 g six hourly for two to four days In children 10–20 mg per pound body weight are given daily in divided doses

Streptomycin may be given intrathecally in doses of 20–100 mg dissolved in 5–10 ml of pyrogen free physiological saline It should be injected into the subarachnoid space slowly For injection into the pleural space 0 5 g may be dissolved in 10 ml of pyrogen free distilled water Streptomycin may be given by inhalation by means

state that the antibiotic is effective in treating syphilis as many years are necessary before final cure can be pronounced (90 91)

Lymphogranuloma Venereum Chloramphenicol in doses of 3 g every eight hours has produced clinical cure

Granuloma Inguinale Chloramphenicol in doses ranging from 20–40 g given over a period of twelve days heals large lesions If Donovan bodies persist for five to seven days after chloramphenicol is started therapy should be continued until healing is complete (92)

Chancroid Willcox reported 2 cases of chancroid which were successfully treated with chloramphenicol Fluctuant buboes still required aspiration (93)

Surgical Infections Chloramphenicol is valuable in treating infection with gram negative organisms and where mixed infections are found it may be used in conjunction with other antibiotics

Other Infections Chloramphenicol has been found to be of value in treating the following diseases Psittacosis trachoma ulcerative colitis herpes zoster infective hepatitis acute laryngo tracheo-bronchitis in children infantile gastro enteritis in which a serologically specific type of colon bacillus (Bact coli B C T) was found chicken pox mumps infective mononucleosis and relapsing fever

STREPTOMYCIN

Following the discovery of penicillin Waksman a soil micro biologist tested many thousands of soil fungi for antibiotic proper ties He paid especial attention to species which inhibited the growth of gram negative organisms He discovered a substance called streptothricin which had considerable antibiotic activity against gram negative organisms but it proved too toxic for clinical use This was followed by an announcement that streptomycin which is derived from a soil actinomycete *Streptomyces griseus* possessed a bacteriostatic action against many organisms *in vitro* (94) Strep tomycin has since been proved to be a very valuable antibiotic clinically against a wide range of bacteria including Myco tuberculosis

Chemical Composition and Pharmacology Streptomycin is a water soluble base whose salts with mineral acids give approxi mately neutral solutions in water The empirical formula is

combination of para amino salicylic acid with streptomycin in the treatment of tuberculosis is said to prevent or retard the development of streptomycin resistance

Clinical Applications

H Influenzæ Meningitis Although this is a rare form of meningitis the prognosis before the use of streptomycin was very grave Early diagnosis is imperative and Allibone et al (97) stress the importance of doing a lumbar puncture on children with fever and any evidence of meningeal irritation Streptomycin is given by intermittent intramuscular injection and this may be combined with intrathecal therapy Treatment should be continued for at least seven days after the cerebro spinal fluid has become sterile

K Pneumoniæ Pneumonia Infection with K pneumoniæ (Bact friedlanderi) may occur in three ways as an acute primary infection as a secondary invader in pneumonia which is primarily due to some other organism or as a chronic pneumonia Gill (98) has drawn attention to the improvement in prognosis which has resulted from the treatment of these infections of the lungs by streptomycin with or without sulphonamides

H Influenzæ Pneumonia Streptomycin is of value in treating bacterial pneumonia when this organism is playing a part in the infection It may be combined with penicillin therapy with advantage

Urinary Tract Infections Streptomycin is excreted in the urine when given by intramuscular injection It is more active in an alkaline urine so that alkalinisation of the urine by giving pot citrate or sod citrate gr 15 four hourly is advisable Streptomycin is of value in treating acute and chronic urinary infections due to Bact coli Staph pyogenes Str fæcalis or K pneumoniæ (Bact friedlanderi) It is often of value in treating infections with Proteus vulgaris and Pseudomonas pyocyanea but these organisms may rapidly become streptomycin resistant The dose is 1–3 g at six hourly intervals for five to seven days or longer if the organism is still sensitive (99 100 101)

Septicæmia Streptomycin is of value in treating septicæmic conditions due to gram negative bacilli Daily intramuscular injections of 2 g of streptomycin for twelve days are recommended

of a suitable nebuliser 0 5 g being dissolved in 10 ml of sterile physiological saline and 1 ml is inhaled every two hours Streptomycin (0 01 g in 1 ml of sterile physiological saline) may be used as eye drops Hourly instillations should be used

Toxicity Toxic reactions to streptomycin appear most frequently when it is used for long periods as in the treatment of tuberculous infections As the commercial preparations have become purer toxic reactions have become less frequent The most important toxic effect of streptomycin is its effect on the vestibular apparatus which is manifest by giddiness and may be accompanied by nystagmus This effect occurs usually between the third and seventh weeks of treatment but it seldom persists for long after the drug is discontinued except in older patients when it may last for months or even indefinitely Bignall et al (95) reported that some success was achieved by giving antihistamines in preventing the vestibular toxic effects but their results were not conclusive Other toxic effects which may occur are skin rashes nausea vomiting drug fever albuminuria and paraesthesiæ Antihistamines are of value in treating skin rashes and possibly in preventing nausea and vomiting Skin sensitisation may develop through repeated local contact This can be avoided by appropriate precautions such as the wearing of rubber gloves by the nurse

Dihydrostreptomycin is produced from streptomycin by reduction of the streptose aldehyde group to an alcohol group by means of hydrogen in the presence of platinum as a catalyst (96) Dihydrostreptomycin is said to have less effect on vestibular function than streptomycin and to be therapeutically as effective The dose is the same as for streptomycin

Antibiotic Activity Streptomycin is active against gram negative bacilli and gram negative cocci and to a lesser extent against gram positive cocci It is often effective against penicillin resistant organisms

Bacterial Resistance Bacteria acquire a high degree of resistance to streptomycin sometimes very quickly To achieve rapid control of the infection it is advantageous to know the actual degree of sensitivity of the causative organism and to select the dosage accordingly The dosage should be sufficient to attain a concentration of streptomycin in the tissues at least four times as great as that needed to inhibit their growth *in vitro* The

mouth in capsules For adult infections 4 g are given daily and the interval between the dose should not exceed six hours for although the drug remains detectable in the blood for as long as twenty four hours after a single dose its concentration begins to fall off after six hours Bacterial resistance to terramycin has been reported *in vitro*

Toxic Effects The drug may cause gastro-intestinal upset nausea and loose stools being predominant In view of its powerful action upon the intestinal flora of the gut changes in the mucous membranes of the mouth as seen with aureomycin and chloramphenicol may be expected but Linsell and Fletcher did not observe any in their series (110)

Clinical Usage Clinically terramycin has been used successfully in pneumonia septicæmia urinary tract infections amœbiasis brucellosis pertussis Rickettsioses primary atypical pneumonia and post-operative sepsis (111)

The following table gives an indication of the best antibiotics to be used in various diseases and in infections with different types of organisms I = first choice II = second choice III = third choice

It should be emphasised that to get the best results the organisms responsible for the infection should be isolated grown in pure culture and the sensitivity of the particular organism to the various antibiotics estimated The antibiotic to which the organism is most sensitive should then be given in full dosage at the intervals recommended Therapeutic failure may be due to the organism becoming resistant or the presence of a feeding focus of infection which is not being reached by the antibiotic Such feeding foci *e g* collections of pus dead tissue or mechanical obstruction to the normal outflow of the body secretion should be treated surgically

Type of Infection or Disease	Penicillin G	Streptomycin	Aureomycin	Chloramphenicol	Sulphonamides
1 Group A β hæmolytic streptococcal infections	I		II		
2 α hæmolytic streptococcal infections	I	II	I	II	
3 Streptococcus fæcalis	II		I		

Streptomycin finds a place in treating sub acute bacterial endocarditis due to penicillin resistant organisms Treatment must be intensive and prolonged and a constant check must be kept on the sensitivity of the causative organism to streptomycin (102 103)

Tularæmia This disease responds well to streptomycin in doses of 2 g daily Treatment is seldom needed for longer than a week (104)

Plague Streptomycin has been reported of value in treating plague *In vitro* streptomycin in a concentration of 0 5–2 5 μg per ml inhibits the growth of Past pestis (105 106)

Local Sepsis Streptomycin may be used in local sepsis due to organisms within its range and against such organisms if they are penicillin resistant It is used as a powder for insufflation or as an ointment

Infantile Gastro Enteritis Streptomycin is of value in treating gastro enteritis in infants The drug is given by mouth before each feed A total dose of 2 g over seven days is recommended (107)

Gonorrhœa Streptomycin may be used with advantage in the treatment of gonorrhœa A single dose of 0 5 g may cure the disease in the acute stage and it does not mask an accompanying syphilitic infection

Chancroid Hirsch and Taggart report streptomycin to be of value in chancroid (108)

TERRAMYCIN

Terramycin was first reported in America by Finlay et al in 1950 (109) It is derived from a soil actinomycete *Streptomyces rimosus* Terramycin is a pure amphoteric compound which is yellow in colour and has a bitter taste It is sparingly soluble in water but it is readily soluble in solutions of strong acids or alkalis when a salt is formed The acid solution is stable up to thirty days at 0 C

Antibiotic Activity Terramycin is in general highly active against gram positive and gram negative cocci and gram negative bacilli Proteus vulgaris is highly resistant but many strains of *Pseudomonas pyocyanea are sensitive It has been reported as* being effective against brucella and rickettsiæ and its effect on viruses is similar to that of aureomycin

Dosage and Administration The drug is usually given by

Type of Infection or Disease	Penicillin G	Streptomycin	Aureomycin	Chloramphenicol	Sulphonamides
23 Primary atypical (non bacterial) pneumonia			1	1	
24 Psittacosis			1	1	
25 Anthrax	1				
26 Syphilis	1		11		
27 Lymphogranuloma venereum			1	11	
28 Granuloma inguinale		11	1	11	
29 Gas gangrene	1				
30 Diphtheria	1 + anti serum				
31 Actinomycosis	1 + sulphadiazine		11		

Table modified from Long et al *Journ Amer Med Assocn* 1949 141 315

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Type of Infection or Disease	Penicillin G	Streptomycin	Aureomycin	Chloramphenicol	Sulphonamide
4 Pneumococcal infections	1		1	11	
5 Meningococcal infections	1				1
6 Gonococcal infections	1	11	11	11	11
7 Staphylococcal infections	1		1		
8 Brucellosis		11 + sulphadiazine	1	1	
9 Whooping cough			1	1	
10 Tularæmia		11	1	11	
11 Typhoid fever				1	
12 H influenzæ infections		1 + sulphadiazine	1	1	
13 Urinary tract infection					
(i) Bact coli	111	11	1	1	1
(ii) Bact aerogenes		11	1	1	
(iii) Proteus vulgaris		11		1	
(iv) Pseudomonas pyocyanea		1	1	1	
(v) Str fæcalis	11		1		
14 Tuberculosis		1 + PAS			
15 Chancroid		11	1	1	
16 K pneumoniæ (Bact friedlanderi) infections		1	1	1	
17 Salmonellæ infections				1	
18 Bacillary dysentery					1
19 Plague		1			
20 Sub acute bacterial endocarditis					
(i) α hæmolytic streptococci	1		1		
(ii) Str fæcalis	1	11	1		
(iii) Staphylococci	1		11		
(iv) Gram negative bacilli		1	1	1	
21 Trachoma	11		1	1	
22 Rickettsial diseases			1	1	

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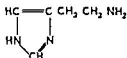
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CHAPTER III

THE ANTIHISTAMINES

IN 1910–11 Dale and Laidlaw (1) demonstrated that the injection of histamine into animals caused symptoms similar to those occurring in anaphylaxis In 1927 Lewis described the effects of intradermal histamine on the human skin and its blood vessels showing that a triple response consisting of capillary dilatation arteriolar dilatation and increased capillary permeability occurred similar to that resulting from minor traumatic damage In 1932 Dragstedt and Gebauer Fuelnegg (2) detected the release of histamine into the circulation of an animal during an anaphylactic reaction thereafter histamine has been identified as the H substance originally postulated by Lewis as responsible for the characteristic anaphylaxis symptom complex When a specific antigen combines with an antibody linked to fixed tissue cells preformed histamine is released from the cells and can be attached to adjacent cells or be carried throughout the body The damage thus caused gives rise to the allergic symptoms in man or to the anaphylactic reaction in experimental animals Most but not all of the symptoms of allergy can be attributed to histamine release for example heparin not histamine is responsible for the increased clotting time of the blood in the anaphylactic state

Properties of Histamine Histamine



has been synthesised and is a white water soluble strongly basic compound Its formation in the body may be by decarboxylation of histidine When injected it has powerful effects on many systems of the body generally due to actions on smooth muscle The more important are as follows — *Skin* When injected locally into the skin marked capillary dilatation and an increase in capillary permeability occur leading to œdema Histamine release also causes itching and cutaneous pain *Cardiovascular system* If there is widespread cutaneous vasodilatation due to circulating histamine

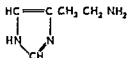
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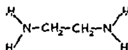
then a secondary fall of blood pressure tachycardia and increase in cardiac output will result *Gastro intestinal tract* Histamine is a *powerful stimulant of the gastric and other secretory glands* and has some stimulatory action on the intestinal musculature *Respiratory system* Histamine has a local action on the bronchioles causing constriction by stimulation of smooth muscle and it also causes an outpouring of mucus In man such broncho constriction does not occur in normals but only in asthmatics (3)

To oppose the effects of an allergic response the following methods might theoretically be adopted —

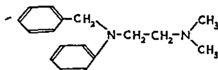
1 Prevent the original antigen antibody reaction 2 Prevent release of histamine from the antigen antibody reaction (antigen desensitisation) 3 Prevent the formation of histamine (by inhibiting histidine decarboxylase *e g* by the use of rutin) 4 Destroy histamine when released (by using a histaminase") 5 Desensitise the tissues to histamine (by repeated injection of small doses of histamine) 6 Use drugs which have actions opposed to that of histamine (*e g* adrenaline which relaxes bronchiolar smooth muscle) 7 Peripherally block the actions of histamine by competitive action at the cell receptors by using drugs which have no direct antagonistic effects It is with this last class of 'anti histamines that we are further concerned

Chemistry of the Antihistamines

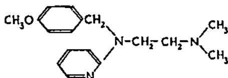
In 1933 Bovet and his co workers at the Pasteur Institute were studying the spasmolytic effects of a number of compounds including 929 F, of which the anti anaphylactic properties were first described in 1937 (4) in the same year the anti adrenaline compound 933 F (piperidino methyl 2 benzodioxane) was also found to be a histamine antagonist (5) Since that date an enormous number of anti histamine compounds have been manufactured and used for clinical or experimental purposes compounds varying in potency and toxicity but in general having the same range of action Many of the antihistamines are derivatives of ethylenediamine



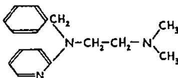
Ethylenediamine



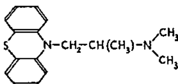
Antergan



Anthisan
(neoantergan)
mepyramine

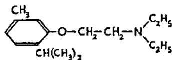


Pyribenzamine
tripelennamine



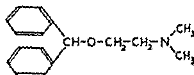
Phenergan
promethazine

Whilst the structures of some other of the important or commonly used compounds are as follows —

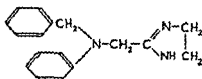


929 F

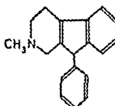
* *G.M C approved name*



Benadryl
diphenhydramine *



Antistin
antazoline



Thephorin
phenindamine *

* G M C approved name

Pharmacological Actions of the Antihistamines

These drugs are readily absorbed from the gut and reach a peak blood level in a few hours (depending on the compound) degradation occurs mainly in the liver and excretion in the urine

Primary Actions This series of drugs antagonises in man all the effects of histamine except the stimulation of hydrochloric acid formation in the stomach *Anaphylaxis* The antihistamines will both prevent and diminish the effects of anaphylactic shock in animals Friedlander et al (6) discuss the relative actions of different antihistamines *They are without effect however on the* phenomenon of the Arthus reaction as this is not due to histamine release unless given in very large doses (7) *Skin Vessels* The antihistamines abolish the flare wheal response which results

from damage to capillaries by histamine (dilatation—exudation) suitable control of this effect has been used for the bioassay of antihistamines in man (8) The effect of an antihistamine (promethazine) in inhibiting increases of capillary permeability due to drugs other than histamine (*e g* adrenaline phosgene) has also been described (9) As well as the antivasodilator effect, the drugs also have a primary vasoconstrictor action by these means they inhibit the hypotensive effects of histamine in anæsthetised animals *Gastro intestinal Tract* The antihistamines effectively inhibit the spasmogenic effects of histamine on intestinal smooth muscle but are not generally intrinsically anti spasmodic This action on guinea pig ileum can be used for biological assay of antihistamines (10) *Lungs* The antihistamines inhibit histamine induced bronchospasm and mucosal œdema

Actions not due to Histamine antagonism *Nervous System* Peripherally the antihistamines exert a local anæsthetic action Centrally in man the vast majority of antihistamines have a depressant effect (Thephorin is a notable exception) In animals stimulation is the general result of moderate overdosage *Miscellaneous Effects* Certain antihistamines have been reported to have an anti hyaluronidase action (11) Amongst other properties described are varying degrees of anti acetylcholine sympathomimetic or sympatholytic activity

Therapeutic use of the Antihistamines

The antihistamines like all other new drugs have been tried for the treatment of most diseases known to medical science In the following section only the more important therapeutic uses will be described

The General Allergic Response During therapeutic desensitisation with allergens the antihistamines have been found useful in the control of local and generalised reactions such treatment should however only be used for patients in whom reactions frequently occur If severe allergic reactions (serum sickness) due to abnormal sensitivity occur during treatment with normally non allergic drugs such as insulin liver extract or penicillin then the use of the antihistamines is advised They usually give considerable relief to the affected skin and mucous membranes although the joint symptoms and pyrexia generally respond badly

Allergic Affections of the Respiratory Tract Hay fever and recurrent vasomotor rhinitis are the disorders most commonly treated with antihistamines The response though purely palliative is generally good though long standing vasomotor rhinitis may not improve so satisfactorily The patient s distress is relieved the sneezing and the itching of eyes and nose respond well but the intranasal œdema may require treatment with ephedrine or similar vasoconstrictor drugs

Although allergic cough also responds well asthma—whether occurring independently or in association with hay fever—is little affected by the antihistamines except possibly by their sedative effect

Many exaggerated claims have been made for the efficacy of the antihistamines in the prevention and treatment of the common cold use of these drugs was originally based on the theory that allergy plays a role in the ætiology of coryza In the last few years however many carefully controlled clinical trials have been performed in which the efficacy of the antihistamines has been balanced against that of placebos and present opinion is that the antihistamines are valueless in preventing or altering the duration of an attack of coryza (12 13) In large doses they may give some symptomatic relief The earlier fallacious reports omitted to consider two important factors the differentiation of coryza from allergic rhinitis and the value of any drug (albeit a placebo) in giving subjective symptomatic relief

Affections of the Skin The true characteristic allergic disorders of the skin urticaria angioneurotic œdema and dermographia respond well to antihistamines Itching is the symptom best relieved œdema in the skin responds moderately well (though the associated serum sickness œdema in *e g* the larynx should be treated by ephedrine) The drugs are more effective in prevention and treatment of the acute condition than in treatment of chronic urticaria Other allergic skin diseases respond variably Atopic dermatitis responds inasmuch as the antihistamines reduce itching and have a sedative action thus the patient scratches less and the lesions may improve on their own When used for treating contact dermatitis again the itching is greatly relieved but cure of the condition depends on removal of the sensitising agent

Affections of the Central Nervous System The antihistamines have been found to possess a depressant action on the vomiting

centre and to be useful in the treatment of nausea and vomiting due to a variety of causes—streptomycin therapy pregnancy motion sickness Much publicity has been given to the successful use of Dramamine (a derivative of benadryl) in 100 mg doses in the prevention and treatment of sea and air sickness (14) The final status of the antihistamines as compared with hyoscine derivatives in the prevention of motion sickness is not yet established Antihistamines have been used with moderate success in the treatment of Parkinson s disease (15) A beneficial palliative reduction in rigidity and muscular cramp is reported with little action however on the tremor and akinesia

Differential Action Dosage and Administration of the Antihistamines

The principal actions of the different antihistamines are the same For practical purposes the drugs may be divided into three groups —

(a) Less potent no sedative action *e g* antistin (thephorin)
(b) Potent moderately sedative *e g* anthisan pyribenzamine
(c) Very potent highly sedative *e g* benadryl phenergan

Important exceptions to the general properties are that thephorin has a stimulant not a sedative action and that phenergan is long acting and has a maximum action at four to twenty hours after oral administration as against the maximum action at one to four hours of the vast majority of the antihistamines

It is impossible for the clinician to remember the actions and properties of the innumerable antihistamines at present on the market and he is advised to become familiar with one member of each group to be used as the clinical condition warrants remembering that individual patients may vary greatly in their responses

The drugs are generally given orally in capsular form or as tablets or pills (which should be swallowed whole) parenteral injection is rarely needed Other methods of administration occasionally used and recommended for particular cases are as an ointment (*e g* 2 / pyribenzamine) for atopic dermatitis or as nasal drops or aerosol spray (*e g* 0 5 / pyribenzamine) for hay fever The usual single dose of compounds in groups (a) and (b) is 100–200 mg (here thephorin is an exception because of its stimulant action the

maximum dose advised is 50 mg) the usual single dose of the more potent compounds of group (c) is 10–50 mg When using an antihistamine for the first time the practitioner is strongly advised to consult the leaflet issued by the manufacturers before administering the drug to the patient

The choice of drug to be used in a particular case depends to a large extent on the individual preferences of the physician and should depend on the degree of sedation required and on the individual response of the patient The size and frequency of the dose should depend on the severity of the symptoms the sensitivity of the patient and also on the patient s size and age—to infants give 1/20–1/4 of the adult dose (depending on the weight) to children aged 2–5 give 1/3 of the adult dose to children aged 5–12 give 1/2 of the adult dose Potable Elixirs are available

Some examples of suitable dosage may be suggested —

Mild hay fever 100 mg of antistin twice daily Serum sickness 100 mg of pyribenzamine four times daily Urticaria with itching 25 mg of phenergan every night Parkinsonism 50 mg of benadryl four times daily

Further reference to doses which have been employed in various diseases may be found in the reviews cited at the end of this chapter It must not be forgotten that the antihistaminics exert a palliative effect only they do not affect the original lesion which is causing the release of histamine

Toxic Effects of the Antihistamines

Despite the claims of the manufacturers the perfect antihistamine has never been produced and probably never will be—that is a drug which combines the maximum action in opposition to histamine with the minimum of side effects In general the more potent the drug the higher its toxicity Toxic side effects so severe that administration of the antihistamine must be stopped occur in about 5% of cases

The principle side effect of the antihistamines is sedation As already described this sedation may be of value therapeutically but in the majority of cases in the ambulant patient at best an unpleasant lassitude is produced and at worst there is a marked degree of diminished alertness drowsiness and detachment from the surroundings With average therapeutic doses 25%–50% of

patients show some symptoms of sedation Benadryl is the most sedative of the common antihistamines and to avoid its soporific effect 5 mg of amphetamine may be given with the benadryl each morning

Conversely excitation convulsions and delirium may occur after very large doses of an antihistamine but need not be feared with the normal therapeutic doses Symptoms due to action on the gastro-intestinal tract are not uncommon epigastric distress nausea or mild colic may occur especially in patients who already have some organic gastro-intestinal disorder Other side actions are dryness of the mouth or palpitation and tachycardia very occasionally skin rashes (occurring with systemic (16) or local (17) administration) and rare cases of agranulocytosis have been reported (18)

Deaths may occur from accidental overdosage this has occurred especially in children and fatalities have been reported (19) of infants or children who ate 500 to 1 500 mg of various antihistamine tablets (presumably thinking that they were sweets (20)) and who died in convulsions within the hour Deaths may occur indirectly usually in adults from motor car or similar accidents which result from the common side effects of drowsiness causing loss of alertness

So it can be seen that the antihistamines are a valuable addition to the therapeutic armamentarium but in a limited field and that their dosage and distribution need to be carefully controlled More detailed reference to the properties and uses of the antihistamines than has been possible in this brief survey may be found in the reviews of Loew (21) pharmacology Huttrer (22) chemistry Hunter and Dunlop (23) general survey Lovelass and Dworin (24) allergy and Feinberg et al (25) general survey

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CHAPTER IV

THE USE OF ISOTOPES IN MEDICINE

ALTHOUGH radioactive isotopes were used in biological investigations by Hevesy (1) Hamilton (2) and Hertz (3) and in therapy by Lawrence (4) before 1940 their extensive use in medicine and related subjects commenced in 1945 Since that time certain nuclear reactors have become available for the cheap production of radioactive isotopes and simple mass spectrometers have been developed for the measurement of stable isotopes Their use in fundamental metabolic studies is now established but their therapeutic application has proved disappointing At the present time only in one disease polycythæmia vera is therapy with a radioactive isotope the treatment of choice

Isotopes of the same element have the same atomic extra nuclear structure and therefore possess similar chemical properties Their nuclear structure and atomic weights differ however and they can be distinguished by physical methods *e g* their atomic weights can be compared by means of a mass spectrometer If the nuclear structure is unstable the isotope is radioactive and a certain proportion of the atoms disintegrate per second emitting particles and radiations which can be detected by such instruments as the Geiger counter or less sensitively by using photographic emulsion The radioactive isotopes can be detected with greater sensitivity than the stable variety and hence can be used at greater dilution in biological experiments They can also be detected *in situ* in the human body and these two advantages have led to their more extensive use in medical research than the stable isotopes However for two important biochemical elements, nitrogen and oxygen the only suitable isotopes are stable

An important unit for the measurement of amount of radioactivity is the curie which represents $3 \cdot 700 \times 10^{10}$ atoms disintegrating per second Smaller units which are usually encountered in tracer work are the milli and micro-curie which are 1/1 000 and 1/1 000 000 of a curie respectively The activity of any particular radioactive isotope decays with time and a measure of this rate of decay is its *half life* which is the constant time interval during which the amount of radioactivity halves in value This may

range from fractions of a second to millions of years for different isotopes If the half life is too short then the isotope may be unsuitable for most investigations e g the oxygen isotope of atomic weight 15 which has a half life of one hundred and twenty-six seconds

Most naturally occurring elements consist of a mixture of stable isotopes e g oxygen consists of 99·757% O^{16} 0·039% O^{17} and 0·204% O^{18} Labelling a group of atoms by means of stable isotopes therefore involves the preparation of material which contains an abnormal proportion of isotopes e g 2% O^{16} 98% O^{18} This abnormal ratio or the degree of dilution of this ratio by the oxygen normally present in the organism or system is detected by the mass spectrometer The use of radioactive isotopes is simpler in principle as only one commonly occurring element potassium contains an isotope which is radioactive Thus it is possible to introduce a group of radioactive atoms all of which are labelled into a system which previously contained no such atoms In practice the group of introduced atoms is not always carrier free but consists of a mixture of radioactive and normally occurring atoms of an element

USE OF ISOTOPES IN METABOLIC STUDIES

The technique of an isotope experiment consists in the introduction of labelled atoms which may or may not be incorporated into molecules into the system under investigation Although the system may contain the same element or compound as the introduced material the labelled atoms are distinguishable because of their physical properties and the movement of these particular atoms can be followed Thus the dynamic properties of a system may be revealed although the chemical state remains in equilibrium

It is not proposed to review in detail here the application of tracers to fundamental metabolic investigations The development of the technique has reached such a stage that it is now best considered in its true place as an invaluable and unique method to be used and considered in conjunction with other methods for the investigation of any particular problem A bibliography of general reference sources for the more recent work is presented at the end of this chapter However it may be of value to state some general considerations which are vital to such applications —

(*a*) The chemical properties of the labelled atoms must be indistinguishable from those occurring naturally That this is approximately the case is shown by determinations of the natural ratio of isotopes in various body compartments Lasnitzki and Brewer (5) have investigated the ratio of K^{41}/K^{39} in biological tissue and found it to be constant except for bone marrow and blood plasma where the ratio is about 3% higher than in other tissue The chemical properties of the isotopes of hydrogen however cannot be regarded as identical The atoms of the isotopes H^3 (tritium) and H^2 (deuterium) have twice and three times the mass of the most abundant naturally occurring atom H^1 This produces differences in their chemical behaviour which particularly affect certain enzyme reactions (6)

(*b*) The radiation from the radioactive atoms must not disturb the normal metabolic processes Ionising radiation may of course even be lethal to living organisms Thus it is advisable to use as small a dose of radioactivity as possible but in general it is not considered that tracer doses cause any short term physiological effect (7)

(*c*) If molecules are to be labelled by the incorporation of tracer atoms then the unambiguous following of such molecules in a chemical process precludes any exchange of the tracer atoms with the corresponding constituents of other molecules That this is generally true has been shown by many workers (8 9 10) However it has been shown that hydrogen atoms bound to oxygen and nitrogen are easily exchangeable with those of water (11) The possibilities of such exchange mechanisms can be explored by multi labelling of a molecule Thus more than one atom of a molecule may be labelled in various positions by different isotopes and the proportion of the isotopes measured by utilising their dissimilar physical properties *e g* by using stable H and radioactive H^3

(*d*) If all the atoms of a labelled element are radioactive then it is generally necessary to introduce only about 10^{-12} *g* of the element in a tracer experiment However it is possible that greater amounts of the element may be necessary because of dilution of the radio active atoms with inert atoms of the same element in the physical production process In the case of compounds the position is less satisfactory because the chemical synthesis will again lower the specific activity (ratio of radioactive to inert atoms) Tracer

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(*b*) The radiation from the radioactive atoms must not disturb the normal metabolic processes. Ionising radiation may of course even be lethal to living organisms. Thus it is advisable to use as small a dose of radioactivity as possible but in general it is not considered that tracer doses cause any short term physiological effect (7).

(*c*) If molecules are to be labelled by the incorporation of tracer atoms then the unambiguous following of such molecules in a chemical process precludes any exchange of the tracer atoms with the corresponding constituents of other molecules. That this is generally true has been shown by many workers (8, 9, 10). However it has been shown that hydrogen atoms bound to oxygen and nitrogen are easily exchangeable with those of water (11). The possibilities of such exchange mechanisms can be explored by multi labelling of a molecule. Thus more than one atom of a molecule may be labelled in various positions by different isotopes and the proportion of the isotopes measured by utilising their dissimilar physical properties, *e.g.* by using stable H^2 and radioactive H^3.

(*d*) If all the atoms of a labelled element are radioactive then it is generally necessary to introduce only about 10^{-12} g of the element in a tracer experiment. However it is possible that greater amounts of the element may be necessary because of dilution of the radioactive atoms with inert atoms of the same element in the physical production process. In the case of compounds the position is less satisfactory because the chemical synthesis will again lower the specific activity (ratio of radioactive to inert atoms). Tracer

experiments demand compounds of high specific activity and negligible mass or the experiment may be carried out under pharmacological rather then physiological conditions Thus the fate of 10 mg of œstradiol in a 100 g rat is of little interest compared with that of 10 μg The metabolism of 14 mg of labelled iodine in humans has been shown to differ very greatly from that of 0 1 μg (12) and Pochin (13) has shown that the metabolism of labelled thyroxine differs with the amount of hormone injected As many isotopes are now available carrier free and detection methods are very efficient the *onus for increase in overall efficiency lies with* the methods of chemical synthesis

Although these considerations and limitations must be carefully considered in isotopic investigations their use in the field of fundamental metabolic studies in conjunction with other methods has brought revolutionary progress in the understanding of the dynamic state of body constitutents This is brought out in the following extract — Components of an animal are rapidly degraded into specific molecular groups which may wander from one place to another The chemical reactions must be balanced so delicately that through regeneration the body components remain constant in total amount and in structure This constancy is not to be taken as an indication that the structural matter of the living organism is inactive and takes little part in metabolism (14 15)

The Use of Isotopes in Clinical Diagnosis

Radioactive isotopes are much more generally used than stable isotopes for clinical diagnosis Even so the number of radio isotopes which can be used is restricted to those with suitable half lives and radiation properties and which can be economically produced (preferably in an atomic pile) The half life must not be inconveniently short and yet must not be so long as to be potentially dangerous to patient and investigators

The isotopes most generally used emit β rays and sometimes also γ rays in addition Even the most energetic β rays which are actually high velocity electrons do not penetrate more than 1 cm of tissue whereas γ rays which could be described as very hard X rays penetrate tissue with very little absorption Thus those isotopes which emit γ rays can be detected *in situ* by Geiger counters but those such as P^{32} Ca^{45} and C^{14} which emit β rays only cannot

be located in any but the most superficial positions It is possible to measure quantitatively the amount of a γ ray emitting isotope in almost any position in the body but even though a pure β ray emitter may be superficially located it is generally impossible to obtain any quantitative results other than a comparative distribution survey The ideal isotope for diagnostic purposes should have a half life of one to fourteen days should emit γ rays of about 0 5 Mev and should be produced economically if possible in a carrier free state by the atomic pile Such an isotope is radioactive iodine I^{131} which has a half life of eight days emits a γ ray of 0 36 Mev as well as β rays and is available at the time of writing in Great Britain carrier free at 10s per 0 5 millicurie from the Harwell pile

The use of isotopes for diagnosis can be divided into two types — static and dynamic investigations

Static Investigations

These investigations are concerned with the distribution of a radioactive isotope with position in a patient at a particular fixed time The survey is usually carried out at a definite time (often

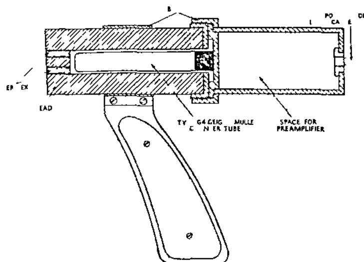


FIG 1 A sectional view of a typical directional Geiger Muller counter The collimating property depends on the lead shielding

experiments demand compounds of high specific activity and negligible mass or the experiment may be carried out under pharmacological rather then physiological conditions Thus the fate of 10 mg of oestradiol in a 100 g rat is of little interest compared with that of 10μg The metabolism of 14 mg of labelled iodine in humans has been shown to differ very greatly from that of 0 1 μg (12) and Pochin (13) has shown that the metabolism of labelled thyroxine differs with the amount of hormone injected As many isotopes are now available carrier free and detection methods are very efficient the onus for increase in overall efficiency lies with the methods of chemical synthesis

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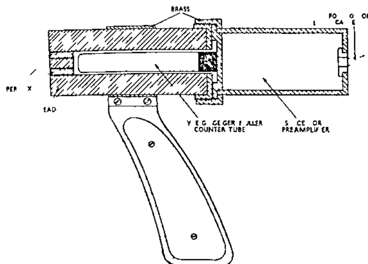


Fig 1 A sectional view of a typical directional Geiger Muller counter The collimating property depends on the lead shielding

twenty four hours) after the administration of the isotope and its distribution localised by means of a specially shielded directional Geiger counter such as is shown in Fig. 1

Tumour Localisation The uptake of an isotope or labelled compound by neoplastic tissue may differ from that of the surrounding tissue. This may yield useful diagnostic information although the differential uptake may not be great enough for therapeutic application. This property of neoplastic tissue is thought to be due to the following causes —

(*a*) *Increased Metabolic Activity of the Tumour Tissue* The specific uptake (per cent of isotope administered per g of tissue) of a particular tissue depends on —

(i) *The amount of the chemically identical element or compound present in the tissue*

(ii) The rate of turnover of the element or compound

In the case of rapidly growing tissue such as tumours the second of these factors is increased and the first factor being the same tumours will show a higher uptake of radioisotope than most normal tissue when specific uptakes are measured shortly after administration of the tracer. Thus Kenney et al (36) found that osteogenic sarcoma had four times the specific uptake of P^{32} than had normal bone. However due to the lower total phosphorus content of such tissue as breast carcinoma its specific uptake although higher than that of normal breast tissue is generally below that of normal bone. In addition the specific activity of such malignant tissue will fall below that of liver spleen and sternal marrow which have higher turnover rates. This precludes the use of P^{32} for general therapy of malignant cells except for *certain disorders of the blood*. It has been used however by Low Beer et al (17 18 19) for the detection and estimation of malignancy of breast tumours. Investigation was made forty eight hours after administration of the isotope by *in situ* measurements but as I^{131} emits β rays only its distribution could only be measured in superficial neoplastic tissue and then not quantitatively. Even so these workers found that carcinomata had at least and benign lesions at the most [illegible] greater uptake than normal breast tissue. There was an increased uptake (as much as 150%) in acute inflammations because of increased vascularisation. They claim that using these criteria as a basis for diagnosis the method

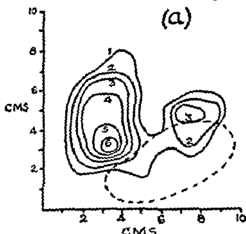
was reliable for 89% of carcinomata 91% of benign lesions and 100% of inflammations

Radiostrontium and radiocalcium are also taken up at high specific activities by osteogenic sarcomata but this property is of little use for diagnostic purposes as both these isotopes are pure β ray emitters It has recently been reported that radiogallium (20) which is selectively absorbed by bone is concentrated to a greater extent by bone tumours This may be especially valuable for diagnosis as radio gallium is a γ ray emitter and is safe because of its short half life

(*b*) *Differential Membrane Permeability of Tumours* Most ions with a negative charge pass more slowly from the plasma into the central nervous system than do other ions This selective barrier is broken down in vessels supplying such abnormal regions as brain tumours abscesses granulomata and traumatised tissues Thus the fluorescein radicle penetrates into such areas more readily than into normal brain tissue and may be detected there during neuro surgical operations by its fluorescence under ultraviolet light irradiation Radioactive di iodofluorescein in which I^{131} is incorporated also exhibits this property and a survey of the skull after administration of this compound to a patient can reveal the presence and location of regions in which the blood brain barrier has been broken down Moore et al (21) report that except for very small tumours with little associated œdematous tissue the method is reliable for the diagnosis and location of glioblastomata ependymomata meningiomata ependymoblastomata and metastatic tumour (in that order of dependability) but is unreliable for acoustic neuromata and astrocytomata It is generally about 95% efficient for diagnosis of the gliomata Ashkenzaky et al (22) who have also done extensive work in this field agree with this figure

(*c*) *Specific Differentiation by Tumour Tissue* It is very desirable that some radioactive compound or element be found that is selectively localised by neoplastic tissue perhaps to be retained there as a synthesised metabolite Such a compound has not yet been discovered which is hardly surprising However the normal thyroid has the ability to absorb one particular element iodine and it also absorbs the radioactive isotope I^{131} very efficiently Thus twenty four hours after oral administration of a carrier free drink containing I^{131} 30% of the dose is taken up by the normal thyroid A survey of the thyroid after such a tracer drink will show the

position of the lobes and isthmus in a normal gland Results of such an investigation are best presented in the form of projection contour maps with lines of equal activity as shown in Fig 2 These maps may indicate to the surgeon areas of high functional activity before thyroidectomy and regions of residual activity after the operation Any disturbance of the normal function of such tissue affects the specific uptake Most thyroid carcinomata have a reduced ability to concentrate radio active iodine and this may be shown in thyroid surveys (Fig 2) As about 5% of cases of well differentiated adenocarcinomata retain this characteristic of I^{131} retention this test can only supply a positive answer regarding the *possible* presence of cyst or tumour Metastases of thyroid primary tumour of a differentiated *type may also* have I^{131} retaining ability and particularly after thyroidectomy these may be detected *and treated by the* use of this radioisotope



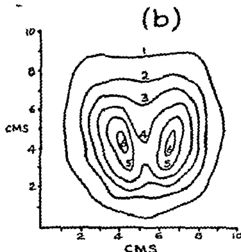


FIG 2 Diagrams of distribution of I^{131} in the thyroid obtained by using a directional counter of the type shown in Fig 1 Figures represent activity (a) Shows the distribution diagram obtained in a particular case of thyroid adenoma The position of the palpable mass is shown by the dotted line It can be seen that this is not composed of functional tissue (b) Shows the type of distribution obtained for a normal gland

Technique of Autoradiography Autoradiography consists in the placing of a tissue section of about 10 μ thickness in contact with a photographic emulsion The presence and location of *radioactive material is indicated by blackening of the emulsion e g* the presence of I^{131} in thyroid colloid can be demonstrated Thus a biopsy followed by autoradiography can be very informative in static investigations A resolution of about 2 μ can be obtained by this method at the present time but it is likely that further advances in technique will lead to its extension from histological to cytological studies

Dynamic Investigations

These studies are concerned with the variation of the quantity of radioistope with time at a particular position

Determination of Distribution Characteristics When a tracer is injected intravenously and its subsequent plasma con centration followed a graph of concentration against time generally shows a multiple exponential decay curve Thus the concentration of radiosodium Na^{24} in plasma shows a rapid decay followed after about four hours by a much more gradual decrease This has been interpreted as a rapid diffusion of the isotope into the extracellular space followed by a more gradual exchange with sodium in such tissue as bone apatite crystal layers and in nervous tissue A similar curve is obtained using radioactive potassium or heavy water (water labelled with deuterium) but for these substances the exponential curve is simple and single The concentration of plasma I^{131} also shows a decay curve but after about twenty four hours the concentration begins to increase This is due to the output of protein bound I^{131} by the thyroid gland which is significant at this time particularly for thyrotoxic patients (23) However in the case of many tracers (Na^{24} K^{4} D_2O) the *concentration in* the plasma is very nearly constant after about twelve hours when allowance is made for excretion Then if it is assumed that the tracer atoms have completely exchanged with the body con stituents the total body content of the introduced compound or element can be found Moore (24) and many others have used D_2O to estimate the total body water of humans He has carried out the same measurement in rabbits and then estimated the water directly finding that the results agree very closely Also just as

inulin is used to estimate extracellular space so tracers can be used to estimate sodium and potassium spaces It should be emphasised that these space values are useful physiological concepts only and represent the effective space occupied by the tracer at the same concentration as in the plasma As many elements are present in intracellular tissue at much greater concentration than in blood plasma the space values do not represent actual space occupied by the element although in the case of sodium this may nearly be so

If R represents the amount of tracer introduced r represents the amount of tracer per ml plasma after equilibrium has been attained and $\bar{\imath}$ represents the amount of substance chemically identical with the tracer in 1 ml plasma after equilibriation then,

$$R/r \times \imath = \text{total body content of the substance and}$$

$$R/r = \text{distribution space of substance}$$

The information which can be obtained by the simultaneous use of heavy water K^{42} and Na^{24} and inulin measurements includes the values for the average intracellular cation concentration Thus heavy water measurements give values of total body water and inulin measures extracellular space Subtraction of these two values gives the intracellular water Use of K^{42} and Na^{24} together with flame photometer measurements of plasma potassium and sodium concentrations gives values for total body sodium and potassium and subtraction of the total extracellular sodium and potassium values (obtained from inulin space × plasma concentration of sodium and potassium) gives the total intracellular potassium and sodium Thus intracellular water potassium and sodium are known and the average intracellular cation concentrations can be calculated Corrections must be made for the amount of tracers excreted

By similar methods total red cell volume and plasma volume can be estimated by the use of tracers Total red cell volume has been estimated by the use of tagged red cells in which radioactive iron (25) or P^{32} (26) or Cr^{51} (27) are incorporated into the erythrocyte Storaasli Friedell et al (28) have used I^{131} tagged human serum albumin to estimate total plasma volume They report that the method has decided advantages over the T 1824 dye method which has previously been used for this estimation a determination which is becoming increasingly important in clinical medicine

The Use of I^{131} in Diagnosis of Thyroid Function

The most frequent application of radioactive isotopes to clinical diagnosis is the study of I^{131} metabolism in selected patients suspected of suffering from disorders of the thyroid gland

I^{131} is a γ and β ray emitter and by the use of suitably designed calibrated Geiger counters (29) the amount of the tracer in the thyroid gland can be estimated *in situ* by external measurements The amount of I^{131} in plasma and urine is usually measured by a β ray counter

If the content of I^{131} in the thyroid expressed as percentage of administered tracer is plotted against time of measurement curves such as Fig 3 are obtained The carrier free I^{131} may be given

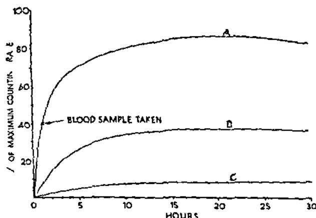


Fig 3 Typical I^{131} uptake curves Curve A was obtained with a thyrotoxic patient A blood sample was taken sixty five minutes after the administration of approximately 15 μc of I^{131} The slope of the curve at this point showed an uptake rate of 0 27 % per minute Assay of the plasma sample showed the I^{131} content to be 0 00041% per ml thus the clearance rate of the gland was 650 ml per minute Curve B shows a typical normal uptake curve and curve C the type of curve obtained with a myxœdematous patient

orally or intravenously for apart from a slight initial delay in uptake after oral administration the results are similar As can be seen from Fig 3 thyroid glands of different activity show differences in the slope of their uptake curves and certain characteristics

of such graphs may be chosen as an index for diagnosis of thyroid function The curves can be described by three characteristics — Initial rate of uptake maximum value of uptake and shape of curve after twenty four hours Both initial rate of uptake and maximum value of uptake increase as the functional activity of the gland increases For patients with thyroid glands of low or normal activity and with normal renal iodide clearance values the uptake remains constant or increases slowly after twenty four hours but for hyperactive glands the uptake decreases after this time This is due to a high rate of output of protein bound I^{131} by such glands Diagnosis may be based on these three characteristics as follows —

Twenty four hour Thyroid Uptake This index is the value of thyroid uptake expressed as per cent of given oral dose measured twenty four hours after administration It has the virtue of great simplicity but has two main disadvantages — (*a*) The measurement may be made during the downward trend of the curve and so may be much less than the maximum value of thyroid uptake (*b*) It is dependent on renal iodide clearance values and may give false results for patients with renal disorders

Werner Quimby and Schmidt (30) have published observations *on 269 patients using this method They found that euthyroid* states give values of 10–40 / for the twenty four hour thyroid uptake value hyperthyroid conditions values over 35% (mostly over 50%) and hypothyroid conditions low values of less than 10 / There was a small overlap of values for euthyroidism and hyperthyroidism and they found lack of correlation between uptake and B M R values in Hashimoto s disease malignant exophthalmos and general hypometabolic states such as Simmonds disease Addison s disease and anorexia nervosa

Thyroid Clearance This index is based on the initial rate of uptake and corresponds in concept to the renal clearance value It is the minimum volume of plasma cleared of I^{131} by the thyroid per minute and is determined by the rate of uptake (per cent per minute) one to two hours after administration of tracer divided by the plasma concentration (per cent per ml) at that time Hence it is expressed in ml /min This index was first shown to be of value by Pochin Myant and Goldie (31) It has the advantage of being a direct measure of thyroid function independent of renal

functions and is probably the best of the numerous suggested methods based on the radioiodine accumulating ability of the gland. Measurements of thyroid clearance should be made after equilibrium has been approximately attained in body tissues other than the thyroid and before the thyroid commences to secrete I^{131} in organic form. As Pochin (13) has suggested, intravenous injection may be of advantage to ensure rapid equilibration, but measurements made one hour after oral administration of the isotope are usually satisfactory. Iodine medication must not be given in the three weeks before the test and the diet must be reasonably normal in iodine content (fish have a high iodine content).

The following values have been found by various British workers:—

Thyroid condition	Workers and reference	No. of cases	Range of values of thyroid clearances ml/min	Mean value of thyroid clearance	Method of administration
Hyperthyroidism	Pochin (13)*	12	84–3[illegible]0	240	Intravenous
	Pochin (31)	11	198–1390	486	oral
	Macgregor (23)	31	30–560	163	oral
Non toxic goitre	Pochin (13)	12	7–4	25	Intravenous
	Pochin (31)	4	28–100	6	oral
	Macgregor (23)	16	15–59	32	oral
Normal	Pochin (13)	12	9–37	25	Intravenous
	Pochin (31)	8	9–38	16	oral
	Macgregor (23)	12	15–57	19	oral
Hypothyroidism	Macgregor (23)	4	0 4–34	13	oral

* Hyperthyroid cases quoted in (13) are clinically more doubtful than those in (31).

There is no doubt that thyroid clearance rates are a reliable measure of thyroid activity and this index is becoming of general use in Great Britain. The use of this test is of value for examining thyroid function in such conditions as anxiety states, essential hypertension, cardiac disease and chronic alcoholism, in which B.M.R. values are nearly always raised independently of the condition of the thyroid. However, as stated by Keating et al. (32), diagnosis based on radioiodine accumulating function of the thyroid gland is as liable to give false results as relying on B.M.R. values, so that these two tests are supplementary. It should be noted that these results are false only from the standpoint of

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have measured arm to foot circulation times by this method Radiosodium was injected into the antecubital vein and a shielded Geiger counter placed against the sole of the foot recorded the delayed pulse of radiation No valuable correlation was established between circulation time and clinical condition Prinzmetal (40) has also applied this method in a procedure which he terms radio cardiography Na [24] was injected very rapidly into the raised arm and a directional γ ray counter placed over the heart recorded the pulses of radioactivity as can be seen in Fig 4 Prinzmetal claims that diagnosis of certain cardiac conditions can be quickly and surely made by this simple procedure A comparison of the method with electrocardiography ballistocardiography and angio cardiography would be useful

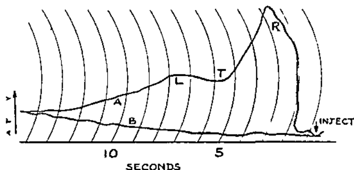


Fig 4 Typical cardiograms obtained by Prinzmetal et al (*Jou n Amer Med Ass cn* 1949 139 617) Curve A was obtained with a normal patient the R wave repres nted blood entering and leaving the right side of the heart whilst the L wave represented blood entering and leaving the left side the trans tion point T being the po nt where most of the blood was in the lungs Curve B was obtained after injecting rad osodium into the antecubital vein of a patient having a superior vena caval obstruction The delayed and ill defined R wave denoted blocking of venous return

An application of isotopes to circulatory conditions which is rather different in principle is the study of build up or rate of disappearance curves ' Quimby and Smith have observed the increase in radioactivity in a particular site after the injection of radiosodium in another part of the body Fig 5 shows such a curve These workers claim that various disorders can be diagnosed by this test and sites of amputation can be decided upon Veall (41)

diagnosis of toxicity and not necessarily from certain other aspects of thyroid activity

Protein bound I^{131} values The estimation of protein bound non radioactive iodine in plasma as a reliable index of toxicity is now realised but the technique of measurement is extremely difficult The concentration of protein bound I^{131} in plasma measured forty eight hours after administration of the tracer is a similar index but the measurement is much easier technically Clark et al (33) McConahey Keating and Power (34) McGregor (23) and Freedberg et al (35) have discussed its value in diagnosis

The relationship of B M R thyroid clearance and protein bound iodine values is not clear at present It seems however that the B M R is a non specific measurement of the rate of body metabolism whereas the thyroid clearance and protein bound iodine values are specifically related to thyroid function Perlmuter Martin and Riggs (36) have shown that although radioiodine accumulating function decreases with age protein bound iodine values remain constant They suggest that this is due to a decreasing rate of utilisation of thyroxine by the peripheral tissues Thus protein bound iodine and thyroid clearance values also represent different aspects of thyroid function Stanbury and Hedge (37) have shown that in certain cases of goitrous cretins the I^{131} accumulating properties may be normal but the protein bound iodine values are very nearly zero Thus there is a blocking of the synthesis of thyroxine in such glands The evidence suggests that I^{131} accumulating function represents the ability of the gland to synthesise iodine to an intermediate compound and is not necessarily a measure of the rate of secretion of thyroxine

Use of Isotopes in Circulatory Studies

The flow of blood may be followed *in situ* by the injection of a γ ray emitting isotope into the blood stream Radioactive sodium (Na^{24} half life 14 9 hours) has been extensively used for this purpose (38) It is not the perfect isotope for the purpose as it rapidly diffuses into the whole extracellular space and it emits a penetrating γ ray which is very difficult to locate directionally However radiosodium can be used without chemical processing it is easily produced and is very safe because of its short half life and general distribution in the body Quimby and Smith (39)

Isotopes have also been used for mechanically tracing the rate of absorption of compounds from various ointments and the extent of the penetration through the skin of injurious compounds such as the nitrogen mustards

RADIOACTIVE ISOTOPES IN THERAPY

Radium or X ray therapy involves the localised irradiation of neoplastic tissue This localisation is carried out by mechanical methods which must necessarily be inefficient and generally if therapy is to be successful the malignant tissue must be more radio-sensitive than normal tissue The possibility of selective irradiation of tumours by radioactive isotopes promised in principle a much more efficient method of therapy However because of the reasons outlined in the section Tumour Localisation only four diseases are now regularly treated by isotopes These are chronic myeloid leukæmia polycythæmia vera functioning metastases of thyroid carcinoma and a non malignant condition thyrotoxicosis The former two are treated with P^{32} and the latter two with I^{131}

Chronic Myeloid Leukæmia P^{32} treatment of this condition results in slightly longer remissions than does X ray therapy An effective dose of 6 millicuries maintained over six weeks will generally result in a remission of over a year (45) Splenomegaly lymphadenopathy hepatomegaly erythrocytopenia and thrombo cytopenia are rapidly improved The advantages over X ray therapy are ease of treatment and lack of radiation sickness

Polycythæmia Vera P^{32} therapy is now the treatment of choice in this condition Six millicuries are generally given intra venously followed one to two months later by a second dose whose magnitude is determined by the individual response Response to treatment is delayed by a time interval corresponding to the mean life of an erythrocyte Complete remission is induced in some patients by a single dose In certain other neoplastic diseases of the hæmopoietic system particularly chronic lymphatic leukæmia P^{3} therapy may be of value Brues and Jacobson (46) have reviewed the comparative therapeutic effects of radioactive and chemo therapeutic methods for such conditions

Thyroid Carcinoma Differentiated metastases of adeno carcinoma of the thyroid may retain I^{131} concentrating ability

states that this test is very dependent on external conditions such as temperature and humidity It would seem that variation between individuals is so great that this test is of the greatest use in the comparative study of the progress of the individual patient with treatment This also applies to a similar method proposed by Kety and Cooper (42) in which the rate of disappearance of radio sodium from an injected site is followed

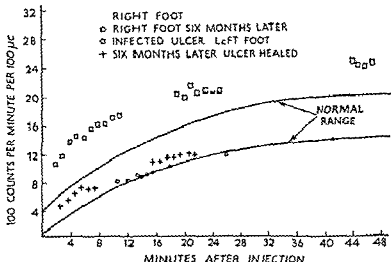


FIG 5 Radiosodium build up curves in a diabetic arteriosclerotic patient with an infected ulcer in the sole of the left foot The upper curve (□) indicates the build up in the left foot in its original condition and the points (•) the build up in the unaffected right foot Six months later the ulcer had healed as a result of local therapy the build up being represented by (+) The right foot build up is unaltered (o) (E H Quimby *Amer Journ Roent genol* 1947 58 740)

Miscellaneous Application of Tracers in Clinical Diagnosis

Further applications of isotopes to clinical diagnosis which are merely in an experimental stage are *the diagnosis of cartilagenous* tumours by investigation with S^{35} labelled sodium sulphate (43) and the use of S^{35} labelled methionine for the study of protein metabolism Kinsel et al (44) state that patients with chronic liver disease idiopathic hypoproteinæmia and Cushing s syndrome showed a significant deviation of protein metabolism from the normal metabolic pattern

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Radioactive Isotopes in Therapy

Radium or X ray therapy involves the localised irradiation of neoplastic tissue This localisation is carried out by mechanical methods which must necessarily be inefficient and generally if therapy is to be successful the malignant tissue must be more radio sensitive than normal tissue The possibility of selective irradiation of tumours by radioactive isotopes promised in principle a much more efficient method of therapy However because of the reasons outlined in the section Tumour Localisation only four diseases are now regularly treated by isotopes These are chronic myeloid leukæmia polycythæmia vera functioning metastases of thyroid carcinoma and a non malignant condition thyrotoxicosis The former two are treated with P^{3} and the latter two with I^{131}

Chronic Myeloid Leukæmia P^{3} treatment of this condition results in slightly longer remissions than does X ray therapy An effective dose of 6 millicuries maintained over six weeks will generally result in a remission of over a year (45) Splenomegaly lymphadenopathy hepatomegaly erythrocytopenia and thrombo cytopenia are rapidly improved The advantages over X ray therapy are ease of treatment and lack of radiation sickness

Polycythæmia Vera P^{32} therapy is now the treatment of choice in this condition Six millicuries are generally given intra venously followed one to two months later by a second dose whose magnitude is determined by the individual response Response to treatment is delayed by a time interval corresponding to the mean life of an erythrocyte Complete remission is induced in some patients by a single dose In certain other neoplastic diseases of the hæmopoietic system particularly chronic lymphatic leukæmia P^{32} therapy may be of value Brues and Jacobson (46) have reviewed the comparative therapeutic effects of radioactive and chemo therapeutic methods for such conditions

Thyroid Carcinoma Differentiated metastases of adeno carcinoma of the thyroid may retain I^{131} concentrating ability

states that this test is very dependent on external conditions such as temperature and humidity It would seem that variation between individuals is so great that this test is of the greatest use in the comparative study of the progress of the individual patient with treatment This also applies to a similar method proposed by Kety and Cooper (42) in which the rate of disappearance of radio sodium from an injected site is followed

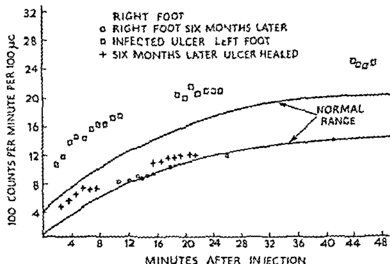


FIG 5 Radiosodium build up curves in a diabetic arteriosclerotic patient with an infected ulcer in the sole of the left foot The upper curve (□) indicates the build up in the left foot in its original condition and the points (•) the build up in the unaffected right foot Six months later the ulcer had healed as a result of local therapy the build up being represented by (+) The right foot build up is unaltered (o) (E H Quimby *Amer Journ Roentgenol* 1947 58 740)

MISCELLANEOUS APPLICATION OF TRACERS IN CLINICAL DIAGNOSIS

Further applications of isotopes to clinical diagnosis which are merely in an experimental stage are the diagnosis of cartilagenous tumours by investigation with S^{35} labelled sodium sulphate (43) and the use of S^{35} labelled methionine for the study of protein metabolism *Kinsel et al (44) state that patients with chronic liver disease idiopathic hypoproteinaemia and Cushing's syndrome* showed a significant deviation of protein metabolism from the normal metabolic pattern





FIG 6 X ray films of the pelvis showing a differentiated metastasis of adenocarcinoma of the thyroid in the right os pub A was taken in June 1949 before treatment and B in January 1950 six months after beginning treatment with 100 mc I^{131} (Green Tait and Worsnop *Brit Journ Radiol* 1951 24 148)

[To face p 7..

They may then be treated after thyroidectomy by the oral administration of large doses of I^{131} (a single dose may be as large as 200 mc) Very dramatic results have been obtained by such therapy (Fig 6) However the proportion of metastases with this ability is small (approximately 10 %) and the value of this method is as a demonstration of principle

Thyrotoxicosis The highly selective uptake of I^{131} by the thyroid gland provides a method of diagnosis and treatment of thyrotoxicosis Radiation of the thyroid by I^{131} is mainly accomplished by the emitted β rays which have a range of only 1 mm in tissue Thus the radiation is very local and even the parathyroids are not greatly damaged Many workers (47) have treated Graves disease by this method which is regarded by a few as the treatment of choice The required dose of 1–5 millicuries I^{131} is determined by the size of the gland the severity of the condition and by the uptake of a preliminary tracer dose of the isotope A possible disadvantage of the method is the theoretical likelihood of a delayed carcinogenic effect but this has yet to be proved although the method is now ten years old However because of this risk most clinicians restrict I^{131} treatment to carefully selected patients

Therapy with radioactive isotopes has not fulfilled its earlier theoretical promise but the work of biochemists in this field may alter the picture The most promising line of investigation seems to be that carried out by Pressman (48) He has been preparing tagged tissue anti sera such as kidney anti serum which on injection is selectively concentrated in kidney tissue The preparation of tagged tumour anti sera is a possible development

Future Development In this field it is safe to prophesy only on future technical developments The most important of these will be the preparation of large numbers of labelled compounds of high specific activity and the development of the scintillation counter This counter has twenty times the efficiency of a Geiger counter for the detection of γ rays and will probably revolutionise *in situ* work

It is to be hoped that the more fundamental metabolic studies with isotopes will be applied in clinical diagnosis not as an interesting novelty but as a unique method of clinical pathology to be used in conjunction with other biochemical procedures

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CHAPTER V

THE VITAMINS

THE actual date of discovery of the vitamins is difficult to place since early travellers even as far back as the Middle Ages were aware that scurvy and similar diseases could be prevented only by feeding with fresh food and more particularly fresh fruit The matter received its first scientific establishment through the work of Sir Frederick Gowland Hopkins and Casimir Funk who proved experimentally that animals could not exist on a diet composed of pure protein fat and carbohydrate Hopkins showed that animals fed on such a diet not only failed to grow but rapidly lost weight and that this state of affairs could be immediately rectified by the addition of small quantities of milk to the diet the quantity added being so small as to make no appreciable difference to the calorie intake Since that time a great deal of work has been done on these accessory food factors as Hopkins originally called them and they have now been separated into a number of groups in all there are to-day some thirty-one vitamins and provitamins which have either been definitely isolated or their presence proved by pharmaco logical experimentation It is proposed now to describe those which are concerned with human nutrition —

Vitamin A This vitamin is present in fats and is soluble in fat solvents It was discovered simultaneously in 1912 by McCollum and Osborn and Mendel These observers showed that the liver of certain fishes such as the cod and animal fats such as butter con tained a substance necessary for the growth of young animals If animals are fed on a diet lacking this vitamin they suffer from a series of degenerative conditions such as xerophthalmia and succumb to intercurrent infections Mellanby has maintained that this vitamin is responsible for raising the general level of resistance against infection and so it has been termed by him the anti infective vitamin It has been shown that a certain chemical test gives a rough indication of the amount of the vitamin which is present in a fat or oil This test consists in treating the oil with a solution of antimony trichloride or arsenic trichloride when a blue colour will develop The amount of vitamin may be judged by the intensity of the blue colour A quantitative estimation may be performed by

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The vitamin has been synthesised and has a potency of 3 to 3 3 million international units per g Vitamin A which occurs in freshwater fish differs only slightly from vitamin A in constitution

Clinical Applications Coward and Morgan (1) give the vitamin A content of foods in international units per g or ml as follows Butter average 60 egg yolk 30 carrots 19 cabbage 9 runner beans 6 milk 3 to 5 In 1937 a technical commission of the League of Nations decided that an adult requires about 3 000 international units of vitamin A daily Children require about 4 000 international units and pregnant and nursing mothers 6 000 to 8 000 international units daily A pint of milk 1 egg 1 oz of butter and a helping of green vegetables will supply about 2 000 international units of vitamin A and 1 000 international units of carotene In war time the average adult only received about 500 to 800 international units of preformed vitamin A daily Carotene is less well absorbed from the alimentary tract in man than is vitamin A probably only 25 % of carotene is absorbed Carrots spinach and broccoli tops are good sources of carotene and with the war time allowance of milk and vitaminised margarine (which has only about half the vitamin A content of butter) ½ lb of carrots should be eaten weekly They are best eaten cooked as they are more easily digested and the carotene is not destroyed by cooking It would appear that the diet of ordinary middle or lower class workers was deficient in vitamin A and to this fact has been attributed the frequency of common ailments such as colds bronchitis and septic complications etc The work of Mellanby has also indicated that lack of vitamin A may play a big part in the ætiology of puerperal sepsis A severe degree of deficiency of vitamin A in the diet leads to xerophthalmia which was prevalent in Denmark in 1917 and is a common cause of blindness in India and China to day Further striking evidence of relative hypovitaminosis A in apparently normal subjects is found by a study of the adaptation of the retina to light It is now known that vitamin A is concerned in the regeneration of visual purple the photo sensitive substance of the retina which is responsible for rod vision Investigations have shown that a large number of apparently healthy persons are affected with night blindness in varying degrees which may be tested by a special photometer Thus Jeghers (2) testing 162 medical students at Boston found 55 deficient in vitamin A as shown by this test Harris and

estimating the colour by means of a Lovibond tintometer The result is expressed as an arbitrary unit which is known as the Lovibond unit if a 20% dilution of the oil in chloroform is used The vitamin may also be estimated by a feeding test in which young rats are given a diet containing the optimum of protein fat carbohydrate and mineral salts and the other vitamins The test is run on a series of animals and as soon as they are losing weight steadily or are ceasing to grow the unknown preparation is administered in varying quantities and the effect noted The relationship between the chemical test and the physiological test is difficult to express since it is highly probable that the efficacy of the vitamin in feeding tests is dependent upon a number of other factors which are at present uncontrolled

As the administration of carotene to animals fed on a diet deficient in vitamin A confers all the benefits they would have received had the vitamin been present it is now generally accepted that carotene is the main precursor of vitamin A being converted in the body into vitamin A and the international unit of vitamin A is taken as 0 6 γ of β carotene From the chemical point of view the relationship is interesting since carotene is a highly unsaturated hydrocarbon with the formula —

```
β Carote   (Karre )
CH   CH
  \ /
   C                  CH                    CH
  / \                 |                     |
CH   C — CH = CH — C = CH — CH = CH — C = CH — CH =
|    ||
CH   C — CH                                                        CH   CH
  \ /                                                                \ /
   CH                  CH                CH                          C
                       |                 |                          / \
        CH — CH = C — CH = CH — CH = C — CH = CH — C    CH
                                                          ||   |
                                                   CH — C    CH
                                                          \ /
                                                           CH
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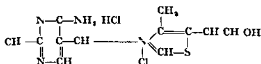
whilst vitamin A has the following formula —

```
CH   CH
  \ /
   C                  CH                    CH
  / \                 |                     |
CH   C — CH = CH — C = CH — CH = CH — C = CH — CH OH
|    ||
CH   C — CH
  \ /
   CH
```

The Vitamin B Complex Vitamin B was originally regarded as a single substance which was necessary for the maintenance of normal health in animals It is now known to be a very complex mixture the following constituents of which have been isolated and synthesised aneurin riboflavin nicotinic acid pyridoxin biotin folic acid *p* amino benzoic acid and pantothenic acid The last two compounds have not yet been proved to play an essential part in human nutrition

Vitamin B_1 The absence of the B_1 factor is responsible for the peripheral neuritis of beri beri which is often accompanied by heart failure and gastro intestinal disturbances The first observations on this interesting substance were made by Eijkman in 1897 He showed whilst working in a prison in Java that fowls suffered from a type of polyneuritis similar to beri beri which at that time affected the prisoners with considerable severity The fowls were fed on polished rice of the same variety as was given to the prisoners and Eijkman showed that when the fowls were fed on unpolished rice *the symptoms disappeared A large scale experiment was carried* out on the prisoners and feeding a carefully controlled group on unpolished rice proved that the disease could be stamped out

Vitamin B_1 is soluble in water In 1932 Windaus (6) determined its composition to be $C_{12}H_{17}N_4OS$ and Windaus (7) and Williams (8) working independently finally established the structure The formula given below is that of aneurin thiamine or vitamin B_1 chloride hydrochloride —



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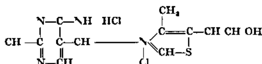
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Abassy (3) reported similar results in school children attending elementary schools in London and Cambridge 40 to 50% were normal 20 to 30% slightly below normal and 20 to 30% definitely deficient A correlation was found between the diet and the dark adaptation tests Among children having ⅔ pint of milk at school 67% were normal on test whereas of those having ⅓ pint or less only 37% were normal Under war conditions in England Yudkin (4) found low adaptation in 9% of children at an urban institution 15% among urban children from poor homes and 18% among some village children It has been shown that the sensitivity to dim light may be greatly increased in these individuals by the administration of vitamin A Yudkin (4) has shown that large doses are necessary to effect prolonged improvement as much as 100 000 international units daily A total dosage of 300 000 to 2 000 000 international units may be required and the beneficial effect may last for weeks or months It is usually given by the mouth in the form of one or other of the fish oils At present the most con centrated form of natural oil is obtained from halibut liver This may be administered either in the form of drops or in capsules An alternative method of administration is to give the various concentrates but it is stated by competent authorities that these are not so stable as the halibut liver oil The preparation Liq vit A Conc (B P Add) *contains 2 500 international units in 1 minim* Cod liver oil contains on an average 2 000 *international units per g and halibut liver oil 160 000 units per g Thus a teaspoonful of cod liver oil contains 6 400 units and a drop of halibut liver oil 3 200 units When administered subcutaneously the preparation is said to be not so active as when given orally Although clinically the risk of hypervitaminosis A seems remote Moore and Wang (5) have produced an experimental condition in rats which they attribute to massive oral doses of vitamin A The symptoms which include tenderness of the limbs and bone fractures are absent if the vitamin is oxidised prior to administration Post mortem examination confirmed the presence of subcutaneous and intra muscular hæmorrhage whilst X ray photographs showed that the majority of the fractures occurred at the centre region of the long bones Although the symptoms in many ways resembled those of scurvy they did not respond to treatment with l ascorbic acid or calciferol*

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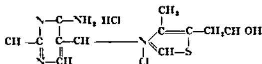
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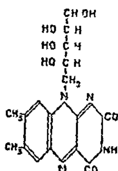
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lips become red along the line of closure and the mucous membrane is thin, shiny and denuded. This is known as cheilosis. Ulceration and fissuring occur at the angles of the mouth resembling the lesions known as perleche, so-called from the verb lecher, as the patient licks the sore with the tip of the tongue. Seborrhoeic accumulations are also noted at the naso-labial folds, on the alae nasi, in the vestibule of the nose and occasionally on the ears and eyelids. These lesions disappeared with the administration of 0·025 mg riboflavin per kg of body weight and reappeared on discontinuing the vitamin. The tongue may also be affected, being purplish red or magenta in colour, clean with flattened or mushroom shaped papillae, and a burning sensation may be complained of. Further, Kruse and his co-workers (27) have found that vitamin B_2 deficiency leads to keratitis and that this condition can be cured both in animals and in human subjects by the administration of the vitamin. Johnson and Eckardt (28) treated thirty-six patients suffering from rosacea keratitis with the vitamin giving 3 mg daily and found that it will correct the condition when it arises from vitamin B_2 deficiency in the diet but not when it arises in patients whose diet is not deficient but who show hypochlorhydria. Engorged limbic vessels are not a definite indication of ariboflavinosis. In order to diagnose the condition on ocular signs new capillaries should be seen by slit lamp microscopy arising from the apices of the limbal loops and spreading on to the cornea around the whole circumference and in each eye. Mann (29) has described a further corneal manifestation of ariboflavinosis with marginal vascularities and opacities in the substantia propria. These changes disappear when riboflavin is administered. Jones et al. (30) described an outbreak of stomatitis due to riboflavin deficiency in a camp in North Africa when the daily riboflavin intake was reduced from 1·61 mg to 1 mg per head. It was quickly cured by giving 100 mg riboflavin in five days or by ½ oz. of fresh yeast daily. Partial syndromes of ariboflavinosis without eye signs or tongue changes have also been described by Duckworth (31) and in one case under our observation a patient complained of a sore hard palate; the mucous membrane ... affected part was pale and the symptoms ... by the administration of 3 to 5 m... of ri... and other remedies having failed

for some of the symptoms often associated with pellagra Its constitution has been revealed through the work on the flavin. It has been known for many years that all plant and animal tissues contain a water soluble yellow pigment known as riboflavin (lacto flavin) which exhibits blue fluorescence This substance has been crystallised by many workers from various sources such as whey egg albumen egg yolk liver urine etc

Gyorgyi Kuhn and Wagner Jauregg (23) suggested in 193 that vitamin B_2 an essential growth factor for rats was a flavin. They were able to show that for finely crystalline lactoflavin a dose of 7 to 10 γ constituted a rat unit The Heidelberg group of workers proved that the synthetic riboflavin is both biologically and chemically identical with the naturally occurring vitamin B_2 The formula of riboflavin is shown here In rats the absence of vitamin B from the diet causes cataract stunting of growth loss of hair and general lowering of condition



Riboflavin

The physiological function of riboflavin like that of aneurin is enzymic in character and is intimately associated with cell respiration As a phosphoric acid ester in association with a protein carrier riboflavin has been identified with Warburg's Yellow enzyme The connection between this important respiratory function and the symptoms of ariboflavinosis are not yet understood

Clinical Applications The chief source of vitamin B_2 is plant tissue particularly young green leaves but milk is a stable and dependable source containing 1 to 2 γ per ml National flour contains 1 3 to 1 9 γ per g and dried yeast 0 1 to 0 2 mg per g

Stannus (24) first described the chief clinical features of riboflavin deficiency twenty years before the discovery of riboflavin Sebrell Butler Wooley and Isbell (25) found by study of the riboflavin balance in ten women that the requirement of this vitamin is approximately 3 mg per day Lack of this vitamin in man produces a condition described by Sebrell and Butler (26) as ariboflavinosis Eighteen women were given a diet with a low riboflavin content and thirteen developed ariboflavinosis

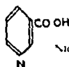
Clinical Applications The distribution of nicotinic acid in foodstuffs has been determined by chemical and microbiological methods Liver kidney pork flesh and yeast are good sources whereas milk contains only small amounts The daily requirement is 15 to 40 mg From 30 to 50 % of the ingested nicotinic acid is normally excreted in the urine as derivatives of nicotinic acid or as trigonelline The blood level and urinary excretion of nicotinic acid in pellagrins does not differ markedly from the normal using chemical methods of assay It has been shown by Kark and Meiklejohn (35) that contrary to the general view porphyrins are not necessarily present in the highly pigmented urine of pellagrins The blue fluorescence of the urine in these cases is attributed to the presence of a substance F_1 of unknown chemical composition A second bluish fluorescent substance F_2 which is possibly thiochrome is a normal constituent of urine and is diminished in quantity in the urine of pellagrins Najjar and co workers (36) found that the administration of nicotinic acid to pellagrins decreased the output of F_1 while raising the quantity of F_2 The minimum effective dosage is still not yet worked out either for the cure or prevention of pellagra but Spies Cooper and Blankenhorn (37) recommend 0 5 g daily given in five doses of 100 mg as safe and effective in the usual case of pellagra For parenteral injection they found that 50 to 80 mg a day in sterile physiologic solution of sodium chloride were effective when injected intravenously Sebrell (38) has emphasised that the best method of prevention is to bring about a permanent change in diet by adding as much green vegetables milk and lean meat as possible Since it seems likely that vitamin B_1 plays some part in the development of the pellagra syndrome the diet must provide for an adequate intake of this factor also

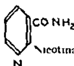
King (39) suggests that Vincent s disease in man in which there is ulceration of the mouth accompanied by the presence of fusiform bacilli and spirochaetes may be a pre pellagrous condition He records good results from the administration of nicotinic acid 250 mg by mouth in water given daily for ten days In glossitis and stomatitis due to nicotinic acid deficiency the tip and sides of the tongue are fiery red and swollen and ulcers form on the tongue and buccal mucous membrane Vincent s organisms may be found in the greyish exudate On account of its vasodilator action intravenous injections of nicotinic acid (5 to 10 mg) have

treatment was omitted Scarborough (32) describing some cases of circumcorneal injection as a sign of riboflavin deficiency comments on this tendency to relapse in certain patients in whom an abnormally large amount of riboflavin appears to be required Manson Bahr (33) *has suggested that vitamin B deficiency* (riboflavin and nicotinic acid) may be responsible for the glossitis found in pellagra sprue pernicious anæmia the nutritional anæmias and idiopathic steatorrhœa Stannus (34) suggests that the first tissue to feel the evil effect of riboflavin deficiency is the capillary endothelium

Nicotinic Acid (Niacin) This is also known as the pellagra preventing (P P) factor Pellagra is a deficiency disease prevalent in districts where maize is one of the staple foodstuffs such as the southern states of the U S A Rumania Italy Egypt and Southern Russia Its symptoms are severe gastro intestinal disturbances and wasting associated with characteristic dermatitis in addition many subjects develop chronic dementia

Experiments on dogs and rats and later on human subjects have shown that pellagra is essentially a deficiency disease which can be cured by the administration of nicotinic acid or its amide Their formulæ are given below —





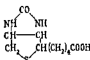
Nicotinic acid like several other members of the vitamin B complex is known as a widespread and important constituent of carbohydrogenase systems Coenzyme I (adenosine ribose diphosphoric ribose nicotinic amide) plays the part of hydrogen carrier to enzyme systems which oxidise such substrates as alcohol lactic acid and malic acid whilst Coenzyme II (adenosine ribose triphosphoric ribose nicotinic amide) plays a similar role in the dehydrogenation of glucose hexose monophosphate and glutamic acid The reduced coenzymes are subsequently reoxidised by transferring their acquired hydrogen to Warburg s yellow enzyme This important function has not yet been related to the clinical symptoms which characterise pellagra

intensify there is a rise in serum iron which runs parallel to the anæmia There is marked hypoplasia of the bone marrow and hæmosiderosis of the liver and spleen Neurological disturbances of an epileptiform character also occur At least in experimental animals all these symptoms are relieved by the administration of pyridoxin There is little doubt that much of the experimental work in the past has been complicated by the failure to recognise the part played by nicotinic acid and folic acid in related conditions

Our clinical knowledge of pyridoxin is far from complete The synthetic compound has been used in substantial quantities in the treatment of anæmias (Vilter Schiro and Spies (50)) muscular atrophy and dystrophy (McBryde and Baker (51)) and Parkinsonism (Spies Hightower and Hubbard (52)) The reports are conflicting and no well defined clinical condition has yet been clearly associated with deficiency The new information regarding the function of folic acid in macrocytic anæmia may help to resolve this problem

Biotin In 1927 Boas (53) showed that rats maintained on a raw egg white diet developed symptoms of an eczema like dermatitis which was associated with a loss of hair Factors which protect rats from egg white injury were isolated independently by Kogl and Tonnis (54) from egg yolk α biotin and by Du Vigneaud et al (55) from liver concentrate β biotin formerly known as vitamin H Despite former assurances that these two compounds were identical it now seems likely that they are indeed distinct (56) The factor responsible for the symptoms of biotin deficiency has been isolated by Eakin Snell and Williams and named Avidin (57) Avidin which has been crystallised (58) is a protein carbohydrate complex of molecular weight about 70 000 It forms a complex with biotin which renders the latter inactive

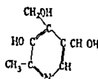
The constitution of β biotin has been elucidated and confirmed by the synthesis by Du Vigneaud and colleagues (59 60) as



The natural vitamin is the *d* compound and as the racemic form tested on rats has only half of the activity of the naturally occurring

been given in cases of cerebral thrombosis by Furtado (40) and in angina pectoris by Neuwahl (41) and in Meniere s syndrome by Atkinson (42) with promising results Nicotinic acid may also be given by mouth in doses of 25 to 150 mg daily according to the *flushing effect produced* in cases of intermittent claudication In some cases the walking capacity of the patient is considerably improved Gottlieb (43) described 5 cases of acute nicotinic acid deficiency characterised by mental confusion or excitement which were relieved by the administration of nicotinic acid in doses of *400 to 600 mg*

Vitamin B_6 (Pyridoxin) In 1934 Gyorgy (44) investigating the condition of acrodynia in rats showed that although the symptoms were associated with a dietary factor none of the known members of the vitamin B complex afforded relief In 1938 five independent groups of workers succeeded in crystallising the essential factor for which the name pyridoxin was finally adopted In the same year degradation and synthesis (45 46) showed that pyridoxin was a pyridine derivative with the constitution shown



Pyridoxin appears to be widespread in the animal and vegetable kingdoms Good sources are to be found in yeast rice polishings and wheat germ and although found in many other foodstuffs it is frequently associated with protein or starch and as such is only assimilated after hydrolysis by cooking

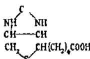
Whilst rats on being deprived of pyridoxin show loss of weight œdema and dermatitis of the extremities the corresponding condition in dogs and pigs is associated amongst other things with a *disturbance* of erythropoiesis Independent workers (Chick et al (47) Wintrobe et al (48) and Borson and Mettier (49)) working variously on pigs and *dogs* agree *that the condition is one of* hypochromic microcytic anæmia The condition although having elements of similarity *with pernicious anæmia does not* respond *to iron* or liver extract therapy On the contrary as the symptoms

intensify there is a rise in serum iron which runs parallel to the anæmia There is marked hypoplasia of the bone marrow and hæmosiderosis of the liver and spleen Neurological disturbances of an epileptiform character also occur At least in experimental animals all these symptoms are relieved by the administration of pyridoxin There is little doubt that much of the experimental work in the past has been complicated by the failure to recognise the part played by nicotinic acid and folic acid in related conditions

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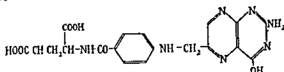
material it is questionable whether the *l* form has any activity (Emmerson (61) Ott (62)) According to Kogl et al (63) the constitution of *a* biotin differs significantly from that put forward by the American school but as the compound has not yet been synthesised no clinical significance is yet attached to it

Biotin is found in close association with other members of the vitamin B complex the chief sources being yeast liver eggs peas and cereals According to Sydenstricker et al (64) the daily requirement of adult humans is about 150 μg

The symptoms of biotin deficiency are not limited to rats As an essential growth factor for many bacteria and moulds (Bios IIB) biotin has been known for over two decades yet the recognition of its clinical importance is of very recent development Experimental deficiency in monkeys may be induced by limiting the biotin intake by adding raw egg white to the diet or by suppressing the synthesis of biotin by the intestinal flora by the administration of sulphonamides (Waisman et al (65)) The syndrome in monkeys is characterised by a thinning of the fur a loss of hair colour and by a conspicuous dermatitis of the face legs and arms all of which symptoms are relieved by the administration of 20 μg of synthetic biotin daily Symptoms of biotin deficiency in man described by Sydenstricker (64) include a scaly dermatitis a greyish pallor of the skin atrophy of the lingual papillae and disturbed erythropoiesis A very remarkable case of clinical biotin deficiency has been quoted by Williams (66)

Folic Acid The subdivision of the vitamin B complex is apparently so endless that it is a pleasant task to be able to report some simplification With the purification of the various reported essential factors it becomes increasingly probable that three factors hitherto treated as separate entities are in fact identical These are *(a) folic acid the acid factor isolated By Williams et* al (67) from spinach and found by him to be a growth stimulant of the organism streptococcus lactis (*b*) the norit eluate factor reported by Snell and Peterson (68) in yeast and liver and found to be an essential factor for the growth of lactobacillus helveticus and (*c*) vitamin B_c a dietary factor described by Hogan and Parrott (69) the absence of which is responsible for the symptoms of macrocytic hypochromic anaemia in chicks The earlier claim of Mitchell and Williams (70) that folic acid bore a close resem

blance to xanthopterin a pigment found in butterfly wings seems to be inconsistent with the inability of this compound to relieve the characteristic symptoms of vitamin B_C deficiency in chicks Notwithstanding this fact the close similarity in physical and chemical properties together with the stimulatory effect upon lactic acid organisms held in common by these three factors support the claim to identity (Pfiffner et al (71)) Some clarification of this problem may be anticipated following the recent characterisation of the liver factor Angier et al (72) have shown by degradation and synthesis that this factor possesses the following structure



N [4 {[(2 amino 4 hydroxy 6 pteridyl) methyl]amino}benzoyl]glutamic acid

The presence of glutamic acid and *p* aminobenzoic acid residues in the molecule is interesting Hitherto the pteridyl ring has not been encountered as such in the biological field

The synthetic compound stimulates the growth of L Helveticus (casei) and is active against anæmia in chicks An analogous synthetic compound prepared by omitting the glutamic acid residue fails to do either but stimulates the growth of Streptococcus fecalis R Angier and co-workers also describe a second factor isolated from a fermentation mixture which as it can be readily converted into the above compound must be closely related to it

Folic acid as its name implies is found in green leaves Concentrates of the vitamin are usually obtained from spinach tops yeast or if an animal source is preferred from liver extracts As this factor is almost certainly synthesised by the intestinal flora it is difficult to estimate the daily requirements Established deficiency in adults has been cured by the administration of 5 mg per day for ten days

Rats maintained on a diet supplemented with aneurin riboflavin pantothenic acid pyridoxin nicotinic acid choline and biotin but containing 1 % sulphaguanidine developed symptoms of agranulocytosis and leucopenia which responded promptly to the administration of crystalline folic acid (Daft and Sebrell (73))

Within four days the white cell counts rose from 2 700 to 14 400 per cubic millimetre with a simultaneous increase in the percentage of granulocytes from 1 to 40

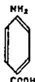
That folic acid not only stimulates the production of white cells but also exerts a profound effect upon erythropoiesis is apparent from the clinical reports of Spies (74 75) on the treatment of nutritional macrocytic anæmia Out of 42 cases of macrocytic anæmia treated 26 responded immediately to treatment with 20 mg of synthetic crystalline folic acid administered either parenterally or orally The cases which included five of Addisonian pernicious anæmia as well as macrocytic anæmia of pregnancy and of sprue showed a sharp reticulocyte response followed by an increase in red cell count and hæmoglobin content Independent confirmation of the efficacy of folic acid in the treatment of sprue has been given by Darby Jones and Johnson (76) who report the successful treatment of 3 classical cases by intramuscular injection of 15 mg of folic acid

The use of folic acid or of synthetic pteroylglutamic acid in the treatment of pernicious anæmia is now universally condemned It is true that the initial response is good Thus Wilkinson (77) showed there is a prompt reticulocytosis with a rapid rise in the red cell count hæmoglobin percentage and in some cases the white cell and platelet counts Later increasing amounts of folic acid are necessary to prevent relapse in some cases and finally there may be no response at all Most serious of all is the fact that neurological changes are not relieved by folic acid and in some cases which were originally free from nervous lesions subacute degeneration of the cord may rapidly appear despite a good hæmatological response It seems even that folic acid may precipitate spinal cord degeneration

Wilkinson and Israëls (78) treated 13 patients suffering from pernicious anæmia with synthetic folic acid conjugates pteroyl γ triglutamic acid (teropterin) and pteroyl α diglutamic acid (diopterin) The hæmatological response was satisfactory indicating that the sufferers from pernicious anæmia can hydrolyse and utilise these folic acid conjugates if given in adequate doses

Para aminobenzoic Acid This substance was first recognised as a vitamin by Ansbacher (79) He fed black and piebald rats on a diet which produced greying of the fur a condition known

as achromotrichia When this diet was supplemented daily by 3 mg of *p*-aminobenzoic acid the animals grew normally pigmented fur within a month Ansbacher (79) also showed that the vitamin improved the condition of chicks reared on a heated vitamin K-deficient diet while Sure (80) found that *p* aminobenzoic acid was an essential dietary factor for lactation and reproduction in the rat It is evident that the above results require further confirmation since some workers have failed to substantiate the claims of Ansbacher The formula of *p*-aminobenzoic acid which is one of the B complex vitamins is as follows



The substance has aroused considerable interest in recent years because of its inhibitory effect on the activity of sulphanilamide This inhibition which was demonstrated by Woods and Fildes (81) *in vitro* and by Selbie (82) in mice infected with streptococcus hæmolyticus led Fildes (83) to conclude that *p* aminobenzoic acid is an essential metabolite for bacteria The vitamin appears to be widespread in plants and animals and has been isolated from yeast which has been found to contain 0 5 mg per 100 g Little is known of the effect of a deficiency of the vitamin in human subjects or of the normal daily requirement Sieve (84) however claimed that *p* aminobenzoic acid caused darkening of the hair in 30 cases of human achromotrichia

Microbiological Assay of Members of the Vitamin B Complex In no single sphere of biochemistry have the methods of microbiological assay been more successful than in the estimation of vitamin B factors As with more complex animals selected strains of bacteria are apparently very discriminating in regard to the composition of the medium upon which they grow Although organisms will fail to grow on a medium lacking in one essential factor, yet over a limited range of concentration they respond in an almost linear manner to addition of micro-quantities of the missing factor It is upon this principle that the method of micro

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biological assay rests Factors essential to bacteria include most members of the vitamin B complex and an impressive list of amino acids including all of those essential to man By using pure solutions to produce standard graphs of growth against added factor it has been possible to estimate with a greater degree of accuracy than is possible by chemical methods the concentration of these factors in foodstuffs

The method will doubtless lend itself to the assay of these factors in biological materials In most assays it has been possible to circumvent the technical difficulty of measuring the growth of bacterial cultures by using lactic acid producing organisms and the linear relation between lactic acid production and the concentration of the missing factor has been demonstrated in many cases

Phycomyces Blakesleanus has been used for a number of years in the determination of aneurin (Schopfer and Jung (85)) Less well known but not less accurate is the use of Lactobacillus Helveticus (L casei ε) for the estimation of riboflavin (Snell and Strong (86)) pyridoxin (Carpenter and Strong (87)) biotin (Snell Hutchings and Peterson (88)) and folic acid (Snell and Peterson (68)) For the estimation of nicotinic acid Lactobacillus Arabinosus is the organism of choice (Barton Wright (89) Kent Jones and Meiklejohn (90)) Other organisms which have been employed in similar assays include Neurospora Sitophila (Stokes et al (91)) Streptococcus lactis (Snell Guirard and Williams (92)) and Leuconostoc Mesenteroides (Barton Wright et al (93)) The latter is also recommended for amino acid assays

Vitamin B_{12} The isolation from liver of the highly potent anti anæmic factor known as vitamin B_{12} was accomplished independently and almost simultaneously in 1948 by Rickes et al (94) in America and by Lester Smith (95) in England Rickes et al (94) by successive purifications of clinically active liver fractions isolated in minute amounts a crystalline substance highly active for the growth of the Lactobacillus lactis Dorner and they called this substance vitamin B_{12} Short (96) had shown that the Lactobacillus lactis requires for its growth two unidentified factors one of which (LLD factor) appeared to be related to the activity of commercial liver preparations used in the treatment of pernicious anæmia It was found that vitamin B_{12} was either wholly or

partially responsible for the LLD growth activity observed for liver extracts. Rickes et al were therefore able to use this micro biological method of assay of the various liver fractions isolated instead of the more tedious and somewhat uncertain clinical trials employed by Ungley (97) on material supplied by Lester Smith

Method of Isolation from Liver Lester Smith and his colleagues have given details of their methods of isolation of vitamin B_{12} in a series of publications (95 98 99 100) They used the method of partition chromatography invented by Martin and Synge (101) Smith (100) outlines the process as follows "The method employs a tube packed with an inert porous powder capable of soaking up a considerable proportion of water without appearing damp the material to be separated is applied to this column dissolved in an organic solvent not completely miscible with water and development is combined with the same solvent These columns do not differ in appearance from the familiar adsorption chromatograms using alumina for example but the mechanism of separation is different it depends on differences in the partition of the various substances present between water (held in the pores of the solid) and the solvent instead of upon differences in adsorbability" Purified liver extracts separated into 3 coloured zones yellow below brown above and pink in between Nearly colourless material was present between the pink and yellow zones

Smith (102) says that the method is very complicated involving at least 6 chromatographic procedures in succession interspersed with other steps of purification such as salting out with ammonium sulphate proteolysis precipitation with phosphotungstic acid and partition between aqueous solution and solvent mixtures The chromatographic steps included adsorption in alumina silica and charcoal The active principle was present in two red bands in the chromatogram column Eventually 20 mg of vitamin B_{12} were obtained from a ton of liver Both Rickes et al (103) and Smith (104) showed that the red crystalline B_{12} contained cobalt to the extent of 4% each molecule probably containing one atom of cobalt and three atoms of phosphorus

Cobalt had been shown by Martin (105) to prevent or cure anæmia in ruminants resulting from feeding on pastures deficient in cobalt The use of the cobalt ion alone is ineffective in the treatment of pernicious anæmia

A further advance was made in the preparation of vitamin B_{12} when it was shown by Rickes et al (106) that the streptomyces griseus from which streptomycin is prepared forms in its metabolism fluid vitamin B_{12} identical with that obtained from liver and producing a similar clinical response in pernicious anæmia It appears probable that there are four or more active forms of B_{12} these have been called B_{12a} B_{12b} B_{12c} and B_{12d} Articles on these forms of vitamin B_{12} have been published by Smith (107) Ungley and Campbell (108) and Chalmers (109)

Clinical Applications Vitamin B_{12} is active in very small doses West (110) tested the crystalline substance isolated by Rickes et al in the clinical treatment of pernicious anæmia in relapse He found that in two patients doses of 3 and 6 micrograms respectively produced a rapid increase in the reticulocytes red cells and hæmoglobin Ungley (97) tested clinically the crystalline vitamin B_{12} prepared by Smith on 53 patients suffering from pernicious anæmia most of whom had received no previous treatment and a few who were in a stage of relapse after discontinuing treatment The vitamin was injected intramuscularly as a single dose The doses were graded logarithmically from 1 25 to 160 micrograms The reticulocytes usually began to rise on the second or third day and reached a peak on the fifth or sixth day A good reticulocytosis was not always a precursor of a correspondingly good rise in the red cell count It was found that the increase of red cells in fifteen days was the best criterion of hæmatological response to the vitamin The amount of vitamin B_{12} required to raise the red cell count to over 4 500 000 per c mm varied from 15 to 140 micrograms The bone marrow showed changes from a megaloblastic to a normoblastic picture Soreness of the tongue disappeared within a few days of the injection and allergic reactions were not met with Ungley has also reported on the favourable effects produced by vitamin B_{12} on the neurological manifestations of pernicious anæmia

For an uncomplicated case of pernicious anæmia he recommended an initial dose of 40 to 80 micrograms followed by 20 micrograms weekly for three months and 30 micrograms every three weeks subsequently

In cases showing neurological changes such as peripheral neuritis or subacute combined degeneration larger doses are required such

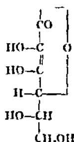
as 40 micrograms every week for six months and 20 micrograms weekly afterwards Berk et al (111) reported rapid regression of neurological manifestations and absence of allergic reactions in a patient sensitive to injectable liver extracts The patient was a forty one year old mulatto woman who had previously been treated with injections of purified liver extracts and folic acid Eight daily injections of 5 micrograms of B_{12} were given On the tenth day of treatment remarkable changes in the neurological signs were noted The numbness and clumsiness of the fingers vanished the plantar responses became flexor the patient could walk without support and vibration sense returned It is clearly essential that the neuro logical changes in pernicious anæmia should be adequately treated before structural damage in the cord renders them irreversible Ungley (112) has also compared the efficacy of vitamin B_{12} with that of more crude liver extracts in the treatment of subacute degeneration of the cord He found that it was equally effective and stated that the existence of a separate neuropoietic factor need no longer be postulated Weekly doses of 40 micrograms of B_{12} are usually effective in the first six months with half this dose for the subsequent maintenance treatment At the least sign of relapse or if there is an intercurrent infection the dosage should be increased

Vitamin B_{12} is available in Great Britain as Cytamen in ampoules containing 20 and 50 micrograms equivalent per ml or Anacobin in ampoules containing 10 micrograms in 1 ml Vitamin B_{12} has also been effective in the treatment of tropical macrocytic anæmia nutritional macrocytic anæmia and sprue It is ineffective in secondary anæmias and in aplastic anæmia

Jacobson and Bishop (113) have brought forward evidence to show that the hæmopoietic effect of vitamin B_{12} is inferior to a comparable dose of commercial liver extract It is suggested that in addition to the primary factor B_{12} accessory factors are required These accessory factors include l tyrosine a peptide xanthopterin tryptophane and guanosine If this is so then a liver extract such as Anahæmin is preferable to B_{12} in the routine treatment of pernicious anæmia

Vitamin C This substance which is water soluble is responsible for protection against scurvy It is found distributed fairly widely in animal tissues and also in the juice of fresh fruit particularly

citrus fruits It has been shown to be *l* ascorbic acid which has the constitution shown below —



The compound is a powerful reducing agent and is unstable in alkaline solution It is estimated biologically by its ability to protect guinea pigs from scurvy and chemically by titration with phenol indo 2 6 dichloro phenol

Clinical Applications The vitamin is administered in the prophylactic and curative treatment of scurvy usually in the form of fresh orange juice Orange juice contains 0 2 to 0 9 mg per ml *rose hip syrup 0 4 to 2 2 mg per ml blackcurrant purée 0 60 to 1 mg per ml* blackcurrant syrup 0 63 mg per ml spinach 0 9 mg per g human milk 0 06 mg and cow s milk 0 02 mg per ml The vitamin C content of cow s milk is lowered by pasteurisation by standing by further heating and by dilution

Vegetables should be cut up only just before cooking and dropped in small quantities into boiling water care being taken that the water does not go off the boil Further if vegetables are not eaten as soon as cooked the ascorbic acid gradually deteriorates *The daily requirement of vitamin C for an adult is 50 to 100 mg* for a child 50 to 150 mg and for an infant 15 to 50 mg Alternatively it may be given as tablets of ascorbic acid each containing 50 mg of crystalline vitamin C 1 to 3 t d s or in adult scurvy ascorbic acid mg 100 to 300 may be administered subcutaneously t i d There is no danger of hypervitaminosis It has been suggested that many chronic illnesses may be due to a degree of vitamin C subnutrition (latent scurvy) Abbasy Hill and Harris (114) have *shown that in active rheumatism the ascorbic acid excretion in the urine is below normal and they conclude that in the infection which underlies rheumatic fever there is a greatly increased metabolic use* of and need for vitamin C and a correspondingly *lower degree of*

saturation of the body reserves Kaiser (115) however determining the vitamin C content of the blood in normal and rheumatic children found no striking difference and considered that vitamin C is probably not a significant ætiological factor in rheumatic infections Thus children may show a fragility of capillaries especially in the early spring Campbell and Cook (116) found that gingivitis characterised by painful and bleeding gums rapidly healed if ascorbic acid was given in doses of 300 mg daily for four days Glazebrook and Thomson (117) made observations on youths between the age of fifteen and twenty years who were having a diet in which the vitamin C was largely destroyed by the methods of cooking and distribution 335 had ascorbic acid administered daily for several months None of them developed pneumonia or rheumatic fever whereas in a group of 1 100 controls there were 17 cases of pneumonia and 16 of acute rheumatism Lack of healing of wounds (Bourne (118)) and of gastric ulcers in patients undergoing treatment has been ascribed to deficiency of vitamin C in the diet In some *cases of pernicious anæmia a satisfactory response to liver therapy* is not obtained until ascorbic acid is also administered Harris (119) concluded that the addition of extra vitamin C to wartime diets increased resistance to infections promoted healing of wounds and improved weight and height gains in children

The diagnosis of vitamin C subnutrition may be carried out as follows Harris and his co workers (120) have studied the concentration of ascorbic acid in the urine both before and after the administration of test doses of ascorbic acid to human subjects They found that normal individuals excrete significant amounts of ascorbic acid in their urine usually over 13 mg daily The vitamin is derived from the diet If the ascorbic acid is withheld from the diet ascorbic acid is excreted at a steady rate for a time indicating the existence of appreciable reserve stores of the vitamin In general the urinary excretion reaches an equilibrium level proportional to the dietary intake of the vitamin Harris regards a daily excretion below 13 mg as suggestive of vitamin C deficiency The effect of a test dose of 700 mg of ascorbic acid per 10 stones of body weight depends on the state of nutrition of the subject with respect to vitamin C In normal individuals there is a sharp increase in the excretion of ascorbic acid usually on the first day following the test dose A patient is regarded as unsaturated

if the excretion rate does not show a rapid increase above the resting level When dealing with patients with a low reserve of ascorbic acid a better idea of the degree of unsaturation may be obtained if the test dose is administered for several consecutive days and the daily output of vitamin recorded

The chief difficulty of the test is due to the instability of ascorbic acid which oxidises rapidly in air For this reason the urine should be collected in dark stoppered bottles and treated with one tenth of its volume of glacial acetic acid Analysis should be performed at once The night urine should be collected in the same way and despatched to the laboratory for analysis early in the morning The whole of the urine passed *in twenty four hours* must be collected Unless these precautions are taken loss of ascorbic acid will occur and may lead to an erroneous conclusion If the output is under 13 mg per day a deficiency of vitamin C may exist For the test dose about 700 mg of ascorbic acid should be given for every 10 stones of body weight and the urine collected for the following two or three days If a small response results the dose should be repeated on one or two further days in order to distinguish between the milder degrees of deprivation and more severe hypo vitaminosis

Harris and Abbasy (121) have proposed a modified test over a shorter period suitable when the collection of full twenty four hourly specimens may be inconvenient At 9 a m the patient is instructed to urinate This specimen is discarded The patient is then instructed not to urinate until 12 noon if possible when the test specimen representing one eighth of the total day s excretion is obtained and analysed The same programme is repeated on a second and third day in order to establish the resting level After this the test dose of 700 mg of ascorbic acid per 10 stones of body weight is administered on one or more further days In these cases the urine samples are collected during the same afternoon over a three hourly period *e g* 2 to 5 p m and analysed at once

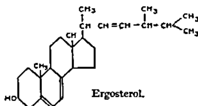
Investigation of the ascorbic acid content of the blood has indicated the existence of a threshold value of about 1 mg per 100 ml of plasma Lower thresholds are sometimes observed Normally the effect of a test dose is to increase the plasma ascorbic acid over the threshold value with the appearance of the vitamin in the urine Ungley (122) showed that with deficiency of vitamin C

the resting level of ascorbic acid in the plasma is low and is only slightly increased by the dose However the plasma value although dependent on the intake of ascorbic acid in the diet is not a good index of a significant deficiency of the vitamin Thus Lund and Crandon (123) in a study of a human volunteer found that the plasma level of ascorbic acid dropped to zero in six weeks whereas clinical scurvy took five months to develop in a subject placed on a diet free from ascorbic acid It was also found that the reducing substance in the buffy coat of centrifuged blood presumably consisting of ascorbic acid took four months to drop to zero and thus provided a more reliable index to the danger of clinical scurvy

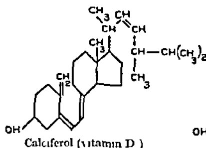
Vitamin D This anti rachitic vitamin is found in fish liver oils but can also be prepared by the irradiation of inactive sterols found in nature Thus the irradiation of ergosterol leads to the formation of calciferol which was formerly thought to be the naturally occurring vitamin The constitution of the naturally occurring vitamin is not certain since many sterols on irradiation produce active compounds At least 10 provitamins D are known the 4 listed below having been isolated in the pure state —

Vitamin D	*Provitamin D*
D	Ergosterol
D_3	7 dehydrocholesterol
D_4	22 dehydroergosterol
D_5	7 dehydro-sitosterol

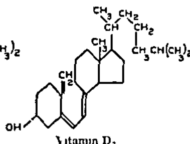
The compound D_3 which has been isolated from tunny fish oil has a high activity when tested in chickens and it is suggested that this is the naturally occurring vitamin D The formulæ for ergosterol calciferol and vitamin D_3 are as follows —



Ergosterol.



Calciferol (vitamin D)



Vitamin D_3

The method of standardising vitamin D consists in employing the same type of method as already recounted for the other vitamins. There is however considerable difference in that the great importance of the presence of this vitamin has succeeded in producing a definite quantitative method of estimation. Rats are fed on a special diet containing no vitamin D until rickets develops. The stock is then divided into groups and to each group graded doses of the preparation are given by mouth. To another series a standard preparation of vitamin D is given and by carefully grading the doses attempts are made to produce the same effect as with the standard preparation and by this means a definition of the activity of the unknown in terms of the standard may be arrived at. The actual technique of determining the degree of healing of the rickets varies considerably. For example X-ray photographs may be taken of the leg bones when uncalcified areas can readily be seen. An alternative method is that known as the line test. In this animals are killed and sections of the long bones are made and the epiphyses are stained with silver nitrate. The preparation is then exposed to the light with the result that at the epiphyseal line a black area is produced. By this means the degree of healing may be studied. It is thus possible to obtain the potency of the unknown in terms of the standard. The international unit is $\frac{1}{10000}$ mg. of irradiated ergosterol. The D vitamins are stable to cooking.

Clinical Applications. The vitamin D content of certain foods according to Coward and Morgan (1) is as follows, expressed as international units per g.: Egg yolk 1.5 to [illegible], cream 0.5, butter 0.4 to 4, milk 0 to 0.1, cod liver oil 480 units per teaspoonful and halibut liver oil 48 units per drop. Fresh herrings contain 250 international units per oz. The use of vitamin D is very wide and

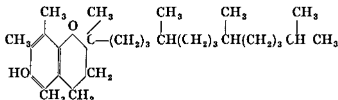
it is usually given either in the form of cod liver oil halibut liver oil or some similar preparation or alternatively as irradiated ergosterol As a prophylactic against rickets most foods given to children are reinforced either by the addition of irradiated ergosterol or by the irradiation of the actual product infants usually require 700 to 1 500 international units daily and as a curative dose 3 000 to 5 000 international units daily The British Pædiatric Association recommends 700 international units daily for a baby during the first year This is contained in 8 drops of Radiostoleum 10 drops of halibut liver oil 12 drops of Adexolin 280 drops of cod liver oil compound and 560 drops of 50 / emulsion of cod liver oil (2 drops = 1 minim) Liq vit D Conc (B P Add) contains 1 500 international units in 3 minims Ten thousand units daily would constitute an overdosage resulting in increased density of bone fever etc Massive doses of vitamin D in the form of irradiated ergosterol (300 000 international units or more daily) have been used to raise the blood calcium in tetany and in the treatment of lupus vulgaris rheumatoid arthritis and hay fever Such doses are not devoid of danger Tumulty and Howard (124) record the case of a man given 750 000 international units daily for twelve days to aid the union of a fracture His blood calcium rose to 15 mg per 100 ml the serum phosphorus not falling below 3 8 mg per 100 ml He was seized with nausea abdominal pains and severe vomiting associated with renal failure The vitamin D had been administered in milk in which vehicle it is ten times more potent than in oil

Dihydrotachysterol (A T 10) although possessing only slight antirachitic potency has also been employed to raise the level of blood calcium in idiopathic and post operative (hypoparathyroid) tetany It appears to possess no special advantage over calciferol for this purpose Albright (125) recommends that the urine of patients under treatment with massive doses of calciferol or A T 10 should be regularly tested for calcium by the Sulkowitch reagent This reagent consists of 2 5 g oxalic acid 2 5 g ammonium oxalate and 5 ml of glacial acetic acid dissolved in water to make 150 ml One volume of urine is treated with 1 volume of reagent If no precipitate of calcium oxalate is obtained there is little calcium present in the urine and the serum calcium level is probably from 5 to 7 5 mg per 100 ml If there is a fine white cloud the level of the calcium in the serum is in the satisfactory range If the mixture

looks like milk the danger of hypercalcæmia is present No method has been worked out for the actual isolation of vitamin D from the urine and therefore we have no definite evidence of a sub rickets condition

Vitamin E It has been shown that animals fed on diets deficient in certain vegetable substances can no longer reproduce The fœtus becomes resorbed following placental changes after the twelfth day In the male this deficiency leads to destruction and atrophy of the testes The distribution of vitamin E in nature is widespread cereal grain green leaves legumes and nuts being good plant sources of the material As far as is known wheat germ oil is the richest source and most workers attempting to obtain the pure vitamin from natural sources have used this as a starting material Vitamin E is classified among the fat soluble vitamins and is destroyed by atmospheric oxygen in the presence of alkali

Three substances α β and γ tocopherol have been isolated and found to possess vitamin E activity α tocopherol has been synthesised by Karrer et al (126) and Todd et al (127) and is as active as the most potent vitamin E concentrates The formula of α tocopherol is shown below



β and γ tocopherol possess one methyl group less than α tocopherol and exhibit only half the activity of α tocopherol when tested on rats

Clinical Applications Vitamin E is put up in 3 m capsules (Viteolin) each containing the unsaponifiable matter of 5 g wheat germ oil or as α tocopherol tablets 3 mg (Ephynal) Fresh dried whole wheat germ may also be given by mouth ½ oz twice daily One international unit corresponds with 1 mg of the acetate

Sterility Good results have been claimed by some authors such as Currie (128) in the treatment of habitual and threatened abortion by the administration of vitamin E in the form of wheat germ oil

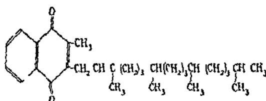
extract 3 m capsules one capsule daily for three to six months up to the onset of labour It is difficult, however to obtain adequate controls and some authorities are sceptical of the results claimed Drummond (129) says that in human beings the process of normal pregnancy like that in the goat and unlike that in the rat may not be dependent on vitamin E

Nervous and Muscular Lesions Bicknell (130) suggests that vitamin E may be deficient in the normal diet of man and that the nervous and muscular systems may suffer in cases of hereditary disposition or from toxic influences such as syphilis He also suggests that the anti sterility factor α tocopherol is not identical with the myotropic and neurotropic factors although all are present in wheat germ oil He treated cases of muscular dystrophy in children and amyotrophic lateral sclerosis and tabes dorsalis in adults with fresh dried whole-wheat germ ¼ oz twice daily for a period up to eighteen months He obtained good results with the myopathies and came to the conclusion that they are deficiency diseases and curable Demole (131) considers that only one factor α tocopherol is needed to prevent paralysis in experimental animals deficient in vitamin E Wechsler (132) obtained good results in two cases of amyotrophic lateral sclerosis treated with pure synthetic α tocopherol in the form of Ephynal tablets 3 mg 2 to 3 t d s Subsequent work by various investigators such as Sheldon et al (133) Doyle and Merritt (134) Denker and Scheinman (135) Ferrebee et al (136) and Fitzgerald and McArdle (137) indicate that vitamin E lack does not cause muscular dystrophies or motor neurone disorders in man Further administration of vitamin E either in the crude form or as the synthetic preparation alone or combined with vitamin B_6 does not cure such disorders This opinion is based both on clinical observations and on the estimation of urinary creatine The excretion of the latter results from nutritional muscular disturbances and if the patient is on a creatine free diet its excretion should diminish as the muscular disease improves Bicknell and Prescott (138) criticise these adverse findings on the efficacy of vitamin E and say that in many cases enormous doses were administered which may act adversely by actually causing a fall in the vitamin E content of the blood They further say that in many of the unsuccessful cases the disease had lasted for many years when recovery of muscular function could hardly be expected

Vitamin K The history of vitamin K the "Koagulations vitamin", dates back to the suggestion of Dam (139) made in 1934 that the spontaneous hæmorrhages found in newly born chicks were the result of the absence of a fat soluble factor from the diet. These hæmorrhages although closely resembling those of scurvy did not respond to the administration of l ascorbic acid and furthermore differed from those of avitaminosis C in that they were associated with hypoprothrombinæmia and a prolonged blood clotting time (Schonheyder (140) Dam et al (141) The analogous clinical conditions in man were recognised by Brinkhous et al (142) in 1937 as hæmorrhagic disease of the newborn and as a hæmorrhagic tendency in cases of biliary obstruction

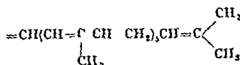
Although fat soluble vitamin K differs from vitamins A and D in that it predominates in plant sources Doisy and his co workers (143) isolated two crystalline compounds vitamin K_1 (alfalfa) and vitamin K_2 (putrefied fish meal) which although similar in physiological activity differed in detail in their chemical composition

Vitamin K_1 has been shown to be 2 methyl 3 phytyl 1 4 naphthoquinone and is represented by the following formula —



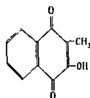
2 methyl 3 phytyl 1 4-naphthoquinone (vitamin K_1)

Vitamin K_2 (144) the second of the naturally occurring vitamins differs in that the phytyl group in the 3 position is replaced by

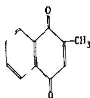


The molecular structure is not very specific and many synthetic 1 4 naphthoquinone derivatives display activity An intact benzene ring and methyl group in the 2 position have been found

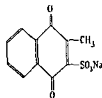
in all active compounds to date Not only can the group in the 3 position be replaced by hydrogen without loss of activity but the quinonoid oxygens may also be replaced by a variety of groups Phthiocol (2 methyl 3 hydroxy 1 4 naphthoquinone) was the forerunner of a series of compounds *in which lyophilic groups* (—OH SO_3H etc) were deliberately introduced to endow the synthetic drugs with water solubility Typical water soluble vitamin K substitutes such as sodium 2 methyl 1 4 naphthoquinone 3 sulphonic acid and 2 methyl 1 4 naphthoquinone bisulphite (Stewart (145)) are readily absorbed even in the absence of bile secretion and are suitable for oral administration (Smith and Owen (146)) The fat soluble analogues are usually administered intramuscularly in an oily medium or orally accompanied by bile salts to aid absorption



Phthiocol



Menaphthone



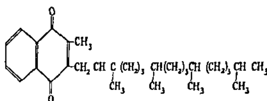
Sodium 2 methyl 1 4 naphthoquinone 3 sulphonic acid

Some doubt exists as to the site and mode of action of vitamin K The older school of thought maintained that the Koagulations vitamin played no direct part in the coagulation of blood but was only associated with prothrombin formation Ample evidence is forthcoming (Fitzgerald and Webster (147) Kark and Souter (148)) that some four hours after the administration of vitamin K to patients suffering from hypoprothrombinæmia there is a distinct rise in the blood prothrombin concentration and a corresponding fall in prothrombin time but as to whether vitamin K is an essential unit in prothrombin or whether it stimulates the formation of prothrombin in the liver no opinion was expressed Lyons (149) has produced definite evidence which bears on this problem According to the classical theory of Morawitz supported by Mellanby (150) *thrombin reacts with* or combines with fibrinogen to form the insoluble fibrin clot Lyons succeeded in dividing

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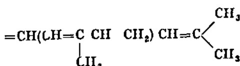
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similar synthetic substance put up in 1 ml ampoules each containing 5 mg for intramuscular injection The synthetic substances have the advantage of being considerably cheaper than the naturally occurring vitamin Bilein capsules each contain gr 5 bile salts

The chief indications for the use of vitamin K are —

1 *Spontaneous subcutaneous and internal hæmorrhages or the liability to bleeding during and after operation* The predisposing condition here is the absence of bile from the intestinal tract resulting in deficient absorption of the vitamin The most important causes are obstructive jaundice biliary fistula and less often acute hepatosis and yellow fever Investigations have shown that vitamin K is a fat soluble vitamin which cannot be absorbed from the intestine in the absence of bile salts The hæmorrhagic tendency which results is due to a low plasma prothrombin level The method for estimating this is described on p 371 and this appears to be the best guide to the need for vitamin K therapy If vitamin K is given by mouth in the absence of bile from the intestinal tract bile salts must also be administered in order to ensure its absorption from the intestine This is not necessary if the vitamin is injected In some cases however absorption appears to be slow after intramuscular injection In cases of operation for obstructive jaundice Klotogen 1 capsule or Kapilon 1 tablet and Bilein 2 gr 5 capsules are given t i d with meals for at least four days before operation and for three to four days subsequently Successful results have been obtained by Macfie et al (153) in cases of obstructive jaundice using intramuscular injections of the synthetic vitamin K Thus Kapilon or Prokayvit 1 to 2 ml may be given daily by intramuscular injection for two to three days prior to the operation and 1 ml subsequently for two to three days In many cases of obstructive jaundice in which the prothrombin level of the blood is low there is a failure of response to vitamin K This is probably due to extensive liver damage In such cases as shown by Kark and Souter (154) if the vitamin fails to raise the prothrombin level the only alternative method of treatment is blood transfusion

2 *Hæmorrhagic disease of the newborn* In newborn infants it is common to find slight hæmorrhagic tendencies during the first week but occasionally the hæmorrhages are so severe as to cause death The investigations of Dam et al (155) indicated that the cause of the hæmorrhage is vitamin K deficiency developing a few

thrombin into two active components Thrombin A and B neither of which alone will convert fibrinogen into fibrin although a combination of both will do so Thrombin A reacts with fibrinogen to form an intermediate compound fibrinogen B between fibrinogen and fibrin Fibrinogen B can be converted into a normal fibrin clot by the addition of Thrombin B or a minute quantity of 2 methyl 1 4 naphthoquinone (vitamin K)

Lyons suggests therefore that either vitamin K is the prosthetic group of the enzyme thrombin or that vitamin K along with a protein carrier acts as an oxido reductase system Support to the former theory is given by the fact that the products of tryptic digest of thrombin give colour reactions characteristic of 1 4 naphthoquinones

Within a few days of birth the danger of hæmorrhagic disease of the newborn recedes and with the establishment of an intestinal flora the child becomes less dependent upon an exogenous source of vitamin K As this synthesis and absorption from the intestine persists throughout adult life it is difficult to get an accurate idea of normal human requirements Sells Walker and Owen (151) place the requirements of the newborn at 1 to 2 μg per day Figures given by other workers are substantially the same

In vitamin K therapy synthetic substitutes have largely replaced concentrates from natural sources Phthiocol (2 methyl 3 hydroxy 1 4 naphthoquinone) the first synthetic analogue no longer finds as widespread use as Menaphthone (2 methyl 1 4 naphthoquinone) which is put up in capsules of 1 to 10 mg for oral administration and in oil for intramuscular injection For more rapid control of hæmorrhage intravenous injection of water soluble derivatives is recommended (Almquist and Klose (152)) The prompt response in these circumstances however is not maintained and a prolonged rise in prothrombin level is best achieved by intramuscular injection of an oil soluble derivative

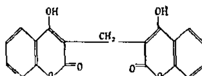
Clinical Applications Various preparations are available for clinical use Klotogen is an oily concentrate of the naturally occurring vitamin It is put in capsules for oral administration Kapilon is an oily solution of a synthetic analogue of vitamin K It is put up in 1 ml ampoules containing 5 mg of the pure substance for intramuscular injection and in tablets of 10 mg and in liquid containing 10 mg in 1 ml for oral administration Prokayvit is a

reduced by 1 6 per 1 000 in the treated infants and recommends the method for general prophylaxis The curative treatment consists in the intramuscular injection of a preparation such as Kapilon 1 ml repeated daily In severe cases blood transfusions should be given

The question as to whether vitamin K deficiency may arise through some dietary lack in otherwise normal subjects cannot yet be answered A study by Kark and Lozner (159) of four non jaundiced subjects with diminished prothrombin levels suggests that there may be a dietary deficiency of vitamin K in man but it seems likely that such a condition can only arise under abnormal circumstances and in conjunction with other nutritional deficiencies A low plasma prothrombin level may occasionally be responsible for hæmoptysis in pulmonary tuberculosis We have had one such case under our care and Sheely (160) reports four cases of hæmoptysis associated with prothrombin deficiency in which the administration of vitamin K appeared to be a likely factor in controlling the bleeding

Readers are referred for further consideration of this subject to the monograph by Butt and Snell (161)

Dicoumarol No account of the relationship of vitamin K deficiency to hypoprothrombinæmia would be complete without reference to the hæmorrhagic symptoms which characterise sweet clover disease in cattle as described by Roderick (162) and Quick (163) The cause of this condition has been shown by the Wisconsin school to be due to the action of a substance present in the sweet clover diet The active principle isolated and synthesised by Link et al in 1941 (164) is Dicoumarol (3 3 methylene bis 4 hydroxycoumarin)



Dicoumarol

Krumer and Barker (165) and Lucia and Aggeler (166) showed that administered orally this compound induces hypoprothrombinæmia *inside seventy two hours unless considerable amounts of* vitamin K analogue are administered simultaneously The fact

days after birth and ceasing usually after the eighth day They have found that in one case the administration of the vitamin together with bile salts through a stomach tube raised the prothrombin level to normal within forty one hours A low prothrombin level however is probably not the sole cause of the hæmorrhage as the level may be low without bleeding occurring Subsequent investigations in America by Sandford et al (156) have thrown some doubt on the matter Although the administration of vitamin K to newborn babies lessened the normal fall in the prothrombin level during the first three days of life hæmorrhagic manifestations occurred as frequently in the babies thus treated as in the untreated These however were all minor hæmorrhages and not those characteristic of hæmorrhagic disease of the newborn They further showed that the administration of vitamin K to the mother before delivery will raise the plasma prothrombin value of the infant Norris and Bennett (157) determined the prothrombin value of the mother and of the newborn child and showed that as there appears to be no relationship between the two it is unlikely that the prothrombin passes directly from the mother to the fœtus and it may be formed in the placenta It does not seem possible to decide by an examination of the maternal blood whether or not vitamin K should be given to the mother before the birth of the child In addition to the pathological hypoprothrombinæmia the clotting time of the blood is prolonged in the hæmorrhagic disease of the newborn The hypoprothrombinæmia occurring in icterus gravis neonatorum anæmia neonatorum and hydrops congenitus also responds to the administration of vitamin K

The prophylactic treatment of neonatal hæmorrhage consists in giving the mother Kapilon 5 tablets or 1½ drams liquid by mouth twelve to four hours before the baby is born and the baby is given 36 drops (18 m) by mouth on the first day of its life Lehmann (158) in Sweden using a vitamin K analogue sodium 2 methyl 1, 4 naphthohydroquinone disulphate which is completely soluble in water and absorbed without the presence of bile salts treated 13 250 full term infants with doses of 1 mg (20 drops of a 0 1% solution) by mouth within an hour of birth the results being controlled by prothrombin estimations In comparison with 17 740 untreated cases Lehmann concluded that death from bleeding was

Vitamin P Szent Györgyi and co-workers (171) isolated from paprika and lemon juice a substance which whilst capable of controlling the petechial hæmorrhages which characterise certain diseases such as vascular purpura was not 1 ascorbic acid Chemical investigation of the material showed that it was an equilibrium mixture of two flavanone glucosides hesperidin and its chalcone eriodictin but as concentrates of the vitamin have been produced by Bacharach and Coates (172) which when tested by these workers were many times more active than crystalline hesperidin the identity of the essential factor is by no means established beyond *doubt*

Clinical Applications Scarborough (173) considered that Szent Györgyi s cases were unsuitable for the demonstration of a new vitamin and that the only suitable cases for investigation are those of known low vitamin intake many of which show capillary fragility unrelated to vitamin C deficiency Scarborough (174) says that as the result of nutritional deficiency and vitamin P lack petechial hæmorrhages occur in the skin with much decreased capillary resistance The patient complains of lassitude with pains in the shoulders and legs but there are no hæmatological changes These symptoms are relieved by the administration of vitamin P but not by vitamin C Scarborough (175) recorded two cases of purpura senilis in which the capillary resistance was low and associated with a tendency to bruise Administration of Permidin apparently increased the capillary resistance and controlled the tendency to bleeding Permidin is available in tablet form for clinical use Each tablet contains the equivalent of 0 25 g of hesperidin the usual dose being 1 to 4 tablets daily Kugelmass (176) treated successfully two cases of allergic purpura in girls characterised by fever abdominal pain bloody stools and purpura He prepared the vitamin P from orange peel The solution contained 50 mg per ml of flavones consisting of eriodictyol glucoside and hesperidin and the dose was 150 mg orally Vitamin P does not appear to be efficacious in the treatment of purpura hæmorrhagica or of mechanical purpura

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that the prolonged clotting time in hæmorrhagic disease is always associated with a fall in blood prothrombin level together with the relatively slow recovery following protection by vitamin K therapy suggests that dicoumarol inhibits the production of prothrombin by the liver Link et al (167) have shown that in rats with pronounced hepatic disorder recovery is very slow even when ample vitamin K is administered

Clinical Applications For the prevention of post operative thrombosis pulmonary embolism and as a prophylactic in thrombophlebitis dicoumarol has advantages over heparin Apart from considerations of cheapness it is effective orally and exercises a more prolonged action The dangers of spontaneous hæmorrhage are never absent however and the course of treatment with dicoumarol must always be controlled by daily determinations of the prothrombin time

The drug is put up in mg 50 tablets called Dicoumarin Allen Barker and Waugh (168) gave to their patients 200 to 300 mg by mouth on the first day and 200 mg on the second day and subsequently it was administered in doses sufficient to keep the prothrombin time between thirty five and sixty seconds (normal fifteen to twenty seconds) This effect should be maintained for about ten days after pulmonary embolism or after the onset of acute thrombophlebitis It should not be used for patients who are bleeding and it is particularly dangerous in subacute bacterial endocarditis Further it should not be used in cases in which there are ulcerating lesions renal disease obstructive jaundice severe liver damage or high fever Zucker (169) reported the successful use of dicoumarol in cavernous sinus thrombosis Davis and Porter (170) recommend the daily administration of three separate doses of 100 mg every eight hours in the treatment of puerperal thrombosis There is usually a delay of twenty four hours after the first dose before the effect is produced and this may persist for two to ten days after the dicoumarol is discontinued Signs of overdosage may occur if the prothrombin time is over sixty seconds They include lassitude aching in the costo vertebral joints bleeding into the skin joints brain viscera or from the gums A blood transfusion using blood not more than four days old will restore the prothrombin time to normal Vitamin K is useless for this purpose

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CHAPTER VI

THE LIVER

A General Consideration of Hepatitis

Progress is being made in the classification of liver diseases on an ætiological and pathological basis and studies over the last ten years both in the human patient and in the experimental animal have led to the discarding of many outworn hypotheses (Himsworth (1))

Acute Parenchymatous Hepatitis Pathologically the lesions in this condition are of two types (*a*) zonal hepatitis (*b*) massive hepatitis

(*a*) The common response of the liver to any mild noxious agent is a centrilobular zonal necrosis the damage being *pan hepatic* such a noxious agent may be chemical *e g* chloroform (Wilcox (2)) micro biological *e g* the virus of infective hepatitis or physical *i e* back pressure in the hepatic veins in heart failure (Boland and Williams (3)) Especially if the disorder is caused by an infection as well as the necrosis there is a periportal inflammatory cell reaction If the damage is slight healing will be complete the centre of the lobules reconstituting from the undamaged peripheral cells this is the common course Repeated attacks of zonal hepatitis may rarely lead to the development of the diffuse fibrosis type of cirrhosis If the attack is severe due to a very virulent infection a massive poisoning or a liver already weakened by dietary deficiency then the lesion will rapidly progress to a massive hepatitis

(*b*) In animals a protein deficient diet alone may be sufficient to cause massive hepatic necrosis (Weichselbaum (4)) in man this probably does not occur The factors responsible are deficiencies of cystine and of tocopherol The pathological lesion is complete destruction of parenchymal cells over large parts of the liver with normal areas (in contrast to the picture in zonal hepatitis) left between the damaged areas In man the condition presents as acute yellow atrophy many of the cases are due to an extension of a severe zonal necrosis T N T and cincophen are poisons very likely to cause massive necrosis (Willcox (2) Himsworth and Glynn (5)) In all these cases malnutrition and protein deficiency

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pathological lesion is a fatty infiltration of the liver cells which if continued over many years will lead to the diffuse fibrosis type of cirrhosis (Rich et al (9)) Many alcoholic cirrhotics show a terminal picture of mixed fibrosis and post necrotic scarring the latter reaction presumably results from the general protein deficiency of the diet

Much attention has recently been paid to Kwashiokor (Waterlow (10)) a complex multiple deficiency syndrome (steator rhœa, anæmia dermatoses) occurring in undernourished children in the tropics A marked feature of the disease is an enlarged fatty liver which likewise may progress to diffuse hepatic fibrosis It is thought that the primary lesion is a pancreatic insufficiency possibly due to protein malnutrition and that the liver disorder and vitamin deficiencies are secondary to this (Davies (11))

Cirrhosis of the Liver

Classification of the clinical disorders known collectively as cirrhosis of the liver is still a matter for argument Three main varieties may be distinguished and mixed types are commonly found

Biliary Cirrhosis Obstruction to the bile passages if continued leads to an ascending cholangitis and cholangio hepatitis with an inflammatory reaction around each portal tract If the infection is not severe enough to cause death or gross parenchymal damage then in the course of years the lesion develops into a periportal unilobular fibrosis without any gross disorganisation of lobular architecture or hyperplastic nodules

Diffuse Fibrosis As an after effect of repeated attacks of zonal hepatitis or of fatty infiltration of the liver from any cause fibrosis develops linking portal and centrilobular tracts Although the fibrosis is apparently unilobular it differs from biliary cirrhosis as the fibrous bands intersect the individual lobules and disorganise the architectural structure Unless complicated by associated sequelæ of subacute hepatitis hyperplastic nodules are absent Clinically the condition may take ten years or more to develop and when discovered (*e g* by the appearance of portal hypertension) is usually compatible with several more years of active life the liver is not grossly enlarged and its surface is only finely granular

Post necrotic Scarring Subacute hepatitis generally progresses to a classical coarse cirrhosis The liver is irregularly intersected

predispose to an extension of the necrotic process Findlay (6) has described an epidemic of infective hepatitis occurring in West Africa in which the mortality rate from massive necrosis was 15 times greater in coloured troops than in white troops the difference being ascribed to discrepancy in nutritional status Acute massive necrosis may likewise occur in pregnancy but in most cases of acute yellow atrophy the causative agent is not known If severe acute massive necrosis kills if less severe the patient will develop a subacute hepatitis complete *recovery of liver structure and of the* patient s health does not occur

Subacute Hepatitis This condition may be compared with the concept of subacute nephritis in that it may result from previous *non fatal attacks of acute hepatitis* (of the massive necrosis *type*) of known or unknown ætiology or it may apparently arise *de novo* In the latter instance there may be a history of severe bacterial infection elsewhere in the body some years previously the condition may occur following pregnancy or the menopause or the patient may have been exposed to an epidemic of infective hepatitis without apparently suffering the acute infection (Barker et al (7)) the virus surviving in the body and wreaking its damage in stealth Inter ruption to the vascular supply of the liver in the initial attack of massive necrosis would leave ischæmic areas more liable to damage by any further noxious factors Patients rarely die in this stage the condition progressing to the post necrotic scarring type of cirrhosis The pathological picture is of a liver showing lesions of every stage normal lobules areas of zonal and of massive necrosis scarring and hypertrophic nodules resulting from previous acute attacks

Dietary Deficiency and Liver Disease It is now considered highly probable that alcohol has no direct toxic action on the liver liver disease in alcoholics being caused by the deficiency in their diets of the lipotropic factors (principally choline and methionine) The principal reasons for this deficiency are a relative poverty (the patient s income being spent on drink) leading to economies being practised by not buying the needed expensive foods of high protein *content* and a gastritis caused by the alcohol diminishing appetite and thus intake of food In rats identical lesions have been produced by keeping the animals on a lipotropic factor deficient diet by feeding them either on alcohol or on sucrose (Best et al (8)) The

there is a state of duodenal catarrh involving the ampulla of Vater with plugging of the orifice of the common bile duct with mucus In 1908 Eppinger (14) described the post mortem findings in a girl nineteen years of age who committed suicide by throwing her self out of a third floor window during an attack of jaundice associated with gastro enteritis The liver was enlarged and icteric and showed typical changes of mechanical jaundice The common bile duct contained dark coloured and very viscous bile The mucous membrane of the stomach showed hæmorrhages The walls of the duodenum were catarrhal and its contents colourless The papilla of Vater was prominent Bile could not be forced through the common bile duct by strong pressure on the distended gall bladder Eppinger thought the obstruction was due to swollen lymphoid tissue near the ampulla of Vater and also at the junction of the cystic duct with the common bile duct

On clinical grounds also certain authorities consider that catarrhal jaundice differs from infective hepatitis for in catarrhal jaundice prodromal symptoms apart from depression are slight, the patient first taking to his bed when the jaundice is manifest In infective hepatitis on the other hand the patient is ill feverish and confined to bed for several days before the jaundice appears

More recently Dible McMichael and Sherlock (15) by aspiration biopsy observations on the liver have found changes in sporadic cases of jaundice identical with those met with in infective hepatitis Further Van Rooyen and Gordon (16) by duodenal intubation studies in 11 cases of infective hepatitis found no evidence of duodeno biliary catarrh These experiments offer no proof that catarrhal jaundice due to primary extra hepatic catarrhal obstruction of the bile ducts is a myth as some would have us believe All they show is that the cases investigated which were assumed to be representative of catarrhal jaundice were in reality examples of inflammatory hepatitis It seems probable therefore that we should not be wise in abandoning Virchow s concept of catarrhal jaundice and that as Boyd (17) writes the basis of catarrhal jaundice may be (1) catarrhal obstruction of the opening of the bile duct (2) toxic necrosis of the liver and (3) cholangitis Providing these lesions are transient the jaundice will be catarrhal in type

Epidemiology Attention was again drawn to outbreaks of jaundice in England by Pickles (18) in 1930 He gave an account

by bands of fibrous tissue separating hyperplastic nodules of regenerated parenchyma from the occasional normal lobule Clinically the condition usually develops within ten years of the initial lesions and once portal hypertension sets in the prognosis is poor The liver is often grossly enlarged and the surface very irregular

Cirrhosis of the Liver in Syphilis In this disease two characteristic types of cirrhosis may be met unrelated to the main classification a diffuse pericellular fibrosis in congenital syphilis and *hepar lobatum* in tertiary syphilis

INFECTIVE HEPATITIS

Relationship to Catarrhal Jaundice Epidemics of jaundice have been described for many years In 1912 Cockayne (12) writing on sporadic and epidemic catarrhal jaundice gave a historical review of epidemic catarrhal jaundice one of the earliest references being to an outbreak in Minorca in 1745 In the British Isles epidemics have been recorded since the Birmingham outbreak in 1852 Each major war has witnessed similar epidemics they were recorded in the siege of Paris in 1870 in the Boer War in Gallipoli during the 1914–18 war and again in the 1939–45 war chiefly in the Mediterranean theatre of operations

Cockayne concluded that simple catarrhal jaundice usually ascribed to gastro duodenal catarrh is except in a comparatively small number of cases the sporadic or endemic form of epidemic catarrhal jaundice He thought that the infection was air borne that the incubation period was short about five days and that there was often a preliminary stage of naso pharyngeal catarrh Cockayne further considered that the liver was probably infected through the blood stream and that inflammation may spread down the bile duct giving rise to catarrhal changes with obstruction This corresponds very closely with the view now held by many that catarrhal jaundice is a sporadic form of infective hepatitis and that in all cases the liver is affected by a blood borne icterogenic agent probably a virus

The evidence however still exists that isolated cases of catarrhal jaundice are due to gastro duodenal or bile duct catarrh and we should not readily accept *ex cathedra* statements to the contrary Virchow (13) in 1865 supported the view that in catarrhal jaundice

changes resembling those of infective hepatitis whereas nine control pigs showed normal livers Cameron (24) injected intra muscularly six volunteers with 1 to 2 ml of whole blood or serum obtained from patients suffering from infective hepatitis in Palestine One man developed jaundice in a month one within two months and the other four within six months As there was infective hepatitis in the district it was not possible to say that all the cases were due to the inoculations Sheehan (25) in 1944 suggested that the icterogenic agent was conveyed by biting insects but this view has not been supported by other workers

Neefe and Stokes (26) recorded an outbreak of infective hepatitis in a summer camp for boys and girls in the Pocono mountains near Philadelphia Out of a population of 573 there were 350 cases It was shown that the water of the girls well was con taminated by fæces from cesspools Healthy volunteers were then inoculated with serum naso pharyngeal washings urine fæces and the infected water It was shown that the icterogenic agent was present in the serum fæces and infected water The serum produced the disease in the human volunteers only when given orally and not when injected parenterally The incubation period was similar to that occurring in the epidemics The agent in the fæces passed through a Seitz filter No evidence was obtained of the transmission of the infection by biting insects It was concluded that it was improbable that the infection was transmitted to the majority of persons by air fomites food milk or directly from infected persons Havens et al (27) in 1945 also showed that the icterogenic agent is present in fæces and that the disease can be reproduced in man by feeding such material in gelatin capsules or by spraying it into the nasal and pharyngeal passages Further jaundice can be produced by feeding serum from cases of infective hepatitis The average incubation period was twenty eight days The ætiological agent probably a virus is in the blood stream before the onset of and during the jaundice It is present in the fæces during the acute stage of the disease and it probably also occurs in the nasal washings It is filtrable and markedly heat stable resisting a temperature of 56 C for half an hour Infection can probably be spread by flies con taminated by infected excreta as is the case with typhoid fever Findlay and Willcox (28) also showed that the oral ingestion of

of 250 cases of epidemic jaundice in Wensleydale The char acteristic features were a pre icteric phase lasting from four to ten days during which the patient suffered from nausea vomiting and pains in the back and limbs This was followed by an icteric stage of about fourteen days Pickles considered that the incuba tion period varied from twenty six to thirty five days that the period of infectivity was short and that the method of spread was respiratory probably by droplet infection In 1942 Newman (19) described an outbreak in the Levant valley in Sussex but he did not hold that the droplet theory of infection was entirely satis factory Fraser (20) in 1931 had brought forward evidence that the disease might be spread by water and further evidence in favour of this view will be referred to later Cookson (21) in 1944 reported an epidemic of 246 cases in Gloucestershire chiefly amongst children attending elementary schools or living in nurseries He rejected the idea that the infection was water borne During the 1939–45 war outbreaks of infective hepatitis occurred especially amongst the troops in Syria North Africa and Italy An interesting account of over 1 000 cases of infective hepatitis amongst American troops in the Mediterranean area was given by Barker Capps and Allen (7) in 1945 The epidemic occurred between February and November 1944

Ætiology It has not been found possible to transmit the disease to animals although Anderson (22) and Hallgren (23) reported the successful transmission of infective hepatitis to the pig but other observers have failed to repeat the experiment It is usually thought that the icterogenic agent is a virus Hallgren (23) in 1942 published an account of epidemic hepatitis in the county of Vasterbotten in Northern Sweden In an outbreak at the Hallnas Sanatorium evidence was obtained that the infection was water borne a corroded sewer having contaminated the well from which the drinking water was obtained Thirty four days after the infected well had been excluded the epidemic came to an end No new cases developed amongst 188 fresh patients who arrived after the exclusion of the well although they were in contact with large numbers of sufferers from infective hepatitis It was found that 32% of those exposed to infection were susceptible A pig was given water from the polluted well and subsequently killed Micro scopical examination revealed necrotic and inflammatory liver

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symptoms there is a low grade pyrexia usually under 100 F and jaundice appears after three or four days (2) Febrile the temperature rising suddenly at the onset to 101 to 103 F Jaundice occurs two to four days later (3) Ambulatory the patient continuing with his work until he is noticed to be jaundiced (4) Hepatitis without any manifest jaundice Hartfall (29) in Malta observed that in 53% the onset was insidious and in 30% it conformed to the febrile type Barker Capps and Allen (7) in the Mediterranean theatre found that amongst American troops in over 80% of cases there was an acute febrile onset the temperature rising to 100 to 103 F associated with malaise headache lassitude anorexia nausea and vomiting The patient complained of chilly sensations rarely of rigors with abdominal discomfort and pains and tenderness in the epigastrium and right hypochondrium Urticaria morbilliform rashes and arthralgia were occasionally met with After three or four days the temperature fell to normal the liver gradually enlarging and becoming tender The spleen was rarely palpable Constipation and diarrhœa were equally common This pre icteric stage was followed five to ten days later by the icteric stage the temperature again rising Nausea and abdominal discomfort were more marked the jaundice lasting from two to four weeks or in some cases for three months or even longer In the average case recovery was complete in from six to eight weeks In this outbreak the stools were rarely clay coloured in other epidemics pasty stools were the rule Cameron (24) in an outbreak in Palestine amongst the troops found the liver enlarged in 97 out of 170 patients and the spleen enlarged in forty six cases Clay coloured stools were found in half the cases in 13 cases the stools were normal in colour in the remainder they were light coloured In fatal cases which are rare the jaundice becomes more intense hæmorrhages may be seen in the skin or there may be pulmonary or intestinal bleeding and in some cases ascites develops The patient becomes irrational and dies in coma

Nervous complications are uncommon but meningitis encephalitis polyneuritis myelitis and upper motor lesions of the pyramidal and striatal type have been described by Lescher (30) Byrne and Taylor (31) and Stokes Owen and Holmes (32) In none of these cases could it be proved that the nervous lesions

fæces or urine derived from spontaneous cases of infective hepatitis will cause typical icteric and sub icteric cases of the disease Seventeen volunteers were given to drink 30 to 50 ml of urine mixed with an equal quantity of milk and 5 developed hepatitis Further 3 g of fæces were ground up in 250 ml of milk and 50 ml were given to each volunteer to drink on an empty stomach Positive results were obtained in 11 out of 47 cases

Alcohol appears to play a part predisposing to infective hepatitis In the Services the disease had a higher incidence amongst officers than amongst other ranks possibly due to the greater amount of spirits consumed by the former Hartfall (29) gives examples of the deleterious effects of alcohol upon an individual who is incubating the disease in provoking an overt attack or in worsening or prolonging an attack otherwise mild and in causing a relapse or second delayed attack of jaundice '

Pathology The disease is probably a systemic infection with a predilection for the liver Dible McMichael and Sherlock (15) studied the pathology of acute hepatitis in 56 cases by aspiration biopsy, and found no histological criteria for differentiating the lesions resulting from epidemic hepatitis arsenotherapy and serum inoculations The cells of the hepatic lobules show evidence of necrosis and autolysis In the areas around the central veins there is rarefaction due to the disappearance of liver cells Small mononuclear cells of the histiocytic class with some polymorpho nuclears and eosinophils tend to accumulate in the portal zones These infiltrations may spread around the periphery of the lobules There is enlargement and hyperplasia of the sinusoidal ' endothelial cells There does not appear to be any fatty change in the cells In over a third of the cases proliferation of the bile ducts was seen in the portal tracts It appears that in the early stages the lesion is chiefly limited to the portal zone with little destruction of the liver lobule The liver inflammation probably begins with the prodromal symptoms Bile thrombi may be seen in the intralobular bile canaliculi In some cases a permanent cirrhosis develop in others there is a mild residual fibrosis which may ultimately completely resolve

Clinical Findings The mode of onset course and severity of the illness are variable Four clinical modes of onset have been recognised (1) Insidious with anorexia and gastro intestinal

Bergh test usually gives a positive direct and indirect reaction falling to normal after about forty days or less

The Urine During the pre icteric stage urobilinogen may or may not be found in the urine with the onset of overt jaundice bilirubinuria occurs

Liver Function Tests Pollock (41) using the intravenous hippuric acid test found that the synthesis was to a large extent correlated with the severity of the attack and the stage of the disease but varied only partly and indirectly with the serum bilirubin concentration The hippuric acid synthesis was noted to be improving while the bilirubin level was still rising and it appeared that liver damage was probably present in the pre icteric period Gordon (42) employing the oral hippuric acid test found evidence of impaired liver function in all of 14 cases examined during the first three weeks of jaundice Barker Capps and Allen (7) observed that the bromsulphthalein test was positive in all cases examined during the first five weeks of the illness and the colloidal gold test was usually positive during the icteric stage Hallgren (23) found that the galactose tolerance test was positive in 12 / of cases Britton (43) using his special technique for the Takata Ara test found a positive reaction beginning within two to three days of the onset of the jaundice the strength of the reaction following closely the severity of the liver damage and the course of recovery

Differential Diagnosis During the pre icteric stage infective hepatitis may be mistaken for influenza gastro enteritis sand fly fever malaria meningitis acute appendicitis enterica group infections or an alchoholic hang over

Treatment *Prophylactic* Gamma globulin obtained from large pools of adult human serum was found by Stokes and Neefe (44) to be a potent agent in the prevention or attenuation of infective hepatitis if injected during the incubation period Gellis et al (45) confirmed this view in two epidemics in the Mediterranean theatre of operation This beneficial effect is possibly due to neutralising antibodies present in the gamma globulin Over 1 700 men were inoculated intramuscularly with 10 ml of globulin and more than 10 000 men served as controls The inoculated belonged to units in which the disease was already rife The incidence of hepatitis in those injected was 0 6 / but of the uninoculated 3 / became infected

were in reality due to the icterogenic agent in all they might have been due to intercurrent causes The death rate from infective hepatitis is very low but in the Mediterranean area it was higher in native than in white troops Relapses are rare but we have seen one patient who had three relapses and died suddenly from hæmorrhage due to ruptured œsophageal varices

Infective hepatitis may not be the innocuous disease that was once though and as described above the sequence of pathological changes leading to one or other form of cirrhosis of the liver has been reported by many workers Wang (33) found that of 447 autopsies of cirrhosis of the liver over a period of sixteen years, 42 could be definitely attributed to previous hepatitis Similarly Rosenak et al (34) elucidated a history of jaundice in 14 out of 106 cases of cirrhosis and Howard and Watson (35) confirm the high proportion compared with a control series Further pathological studies of cirrhosis following hepatitis are reported by Wyllie and Edmunds (36) on 5 fatal cases in children they conclude that it is impossible to tell in the icteric stage which cases will go on to cirrhosis Sherlock (37) stresses the value of liver biopsy as a guide to progesss in a study of 9 cases of cirrhosis following acute hepatitis

Laboratory Findings *The Blood* The white cell count is normal or there is a slight leucopenia A leucocytosis would indicate that the diagnosis is probably incorrect and leucocytosis is a feature of Weil s disease The sedimentation rate of the red cells was found by Miles (38) to be usually normal during the icteric stage of the disease but often raised in the pre icteric period and during convalescence This author suggests that an excess of bile salts in the blood is responsible for the low sedimentation rate during the active phase of infective hepatitis Barker Capp and Allen (7) also reported that the sedimentation rate is usually normal in the presence of jaundice Evans (39) found that the sedimentation rate may remain raised for some weeks after an attack Hoagland and Shank (40) in a series of 16? cases found that the sedimentation rate was *normal* or only slightly raised during the first days of the disease but *frequently* raised to values of 25 to 50 mm per hour during convalescence and gradually falling to normal late in the period of recovery The serum bilirubin is generally normal during the pre-icteric *stage* rising later to levels of 10 mg /100 ml or higher according to the intensity of the jaundice. The Van den

an untreated control series Schemm and Layne (53) found a high fluid intake and an acid ash and low sodium diet valuable in the removal of ascites whilst infusion of salt poor human albumin is not generally recommended (Faloon et al (54)) Rosenak et al (55) used testosterone propionate in doses of 25–100 mg three times weekly this caused a marked improvement in their patients feeling of general well being

Homologous Serum Jaundice

It is believed that homologous serum jaundice results from the introduction of an icterogenic substance into man by the injection of human blood serum or plasma The amount of serum injected may be very small as little as 0 05 ml

As far back as 1885 Lurman (56) reported an outbreak of jaundice following vaccination against small pox with glycerinated humanised lymph Out of 1 289 workers so vaccinated in a ship building yard at Bremen 191 developed jaundice several weeks later whereas 500 others vaccinated with a different batch of lymph were unaffected

We now know that homologous serum jaundice may arise in a variety of ways of which the following are the most important — By the injection of yellow fever vaccine prepared with human serum as the result of prophylactic inoculation with measles convalescent serum or mumps convalescent plasma and following transfusion with whole blood plasma or serum It may also result from the use of syringes contaminated with infected blood and employed either for intravenous intramuscular or subcutaneous injections or for removing blood by venepuncture This latter group includes post arsenical jaundice occurring in veneral disease clinics jaundice following the injection of penicillin or Pentothal and occasional outbreaks of jaundice in clinics for the treatment of diabetes mellitus and arthritis or in sanatoria These varieties will now be considered in more detail

Jaundice following Yellow Fever Vaccination Findlay and MacCallum (57) in 1937 and subsequently drew attention to the occurrence of jaundice following immunisation against yellow fever with a vaccine consisting of yellow fever virus suspended in human serum They observed 89 cases among 3 100 persons immunised over a period of five years the latent period varying

Curative Complete rest in bed from the onset appears to shorten the course of the disease in the majority of cases It is usually advised that the diet should be high in protein (200 g) and in carbohydrate (400 g) and low in fat (40 g) Darmady (46) on the other hand considered that there is no difference in the results of treatment whether a high or a low protein diet is given A light diet with plenty of milk from which the top layer of cream has been removed is usually perfectly suitable During the acute stage the patient should take fluids up to 5 pints a day If this amount cannot be taken by mouth 10% dextrose in water should be given intravenously or if vomiting persists 5% dextrose in saline should be transfused to make up the deficiency If there are hæmorrhages or if the plasma prothrombin content is low vitamin K should be injected intramuscularly in doses of 5 mg daily and for serious hæmorrhage a blood transfusion should be given The patient should take no alcohol during the disease or for a year subsequently Methionine an essential amino acid and a lipotropic factor which promotes the transfer of fat from the liver to the storage depots has been recommended as an aid to treatment but Wilson Pollock and Harris (47) giving 5 g of methionine daily by mouth to alternate patients in a series of 100 cases of infective hepatitis found that the methionine as judged by clinical and biochemical criteria had no significant effect upon the severity or duration of the disease or the incidence of relapses Similar results were obtained by Higgins et al (48) Aureomycin has been tried in the treatment of infective hepatitis It appears to have no curative effect in the acute phase but was reported as being some what beneficial in the treatment of coma or of chronic liver disease (Farquhar et al (49) Shaffer et al (50)) Finally the patient should not be allowed to get up until the temperature is normal, the liver has returned to its accustomed size and is no longer tender there are no symptoms such as lassitude and nausea and the serum bilirubin has been normal for a week

Treatment of the cirrhotic stage is the same as treatment of cirrhosis of the liver of other ætiology—rest and a basic diet (containing ample quantities of the vitamin B complex) are probably still the most important factors (Klatskin and Yesner (51)) Ralli et al (52) report that treatment of cirrhosis for three years with intra venous liver extract resulted in a 44% survival rate against 1-% in

from the Ministry of Health in 1943 (64) as the matter was now considered to be one of major importance It was thought that the jaundice was due to the injection of human blood products and that probably further cases would occur particularly after trans fusion and such indeed has proved to be the case Cockburn et al (65) have reported some cases of homologous serum hepatitis following measles prophylaxis using dried human plasma Ten children were inoculated using a batch of plasma which was subsequently found to be icterogenic Seven of the children developed a severe form of jaundice and three of the seven died One of the fatal cases had symptoms of encephalitis without apparent jaundice

Jaundice following Mumps Convalescent Plasma Beeson Chesney and McFarlan (66) in 1944 reported the occurrence of 101 cases of jaundice following the inoculation of 266 men with mumps convalescent plasma The onset was in the majority of cases between the fifty ninth and ninety fourth day after inocu lation The jaundice was considered to be due to a hepatoxic agent in two batches of plasma

Jaundice following Transfusion Cases of jaundice following at an interval of about three months after blood transfusions were recognised in England in 1942 Morgan and Williamson (66) reported that 9 patients who were treated with large transfusions of plasma or serum 7 suffering from intermittent claudication 1 from diabetes mellitus with gangrene of the toes and 1 from calcinosis circumscripta developed jaundice seven to ten weeks later It was not possible to trace the icterogenic agent to definite batches of serum or plasma The authors stated that the jaundice might be due to a virus or to some toxic chemical substance such as altered plasma protein but there was no evidence to favour one or other of these hypotheses Beeson (68) in America in 1943 reported 7 cases of jaundice occurring one to four months after the transfusion of whole blood or plasma and concluded that isolated cases may not infrequently be met with Steiner (69) also recorded 5 cases of jaundice following transfusions with whole blood or human plasma given to soldiers when they were wounded two to three months previously There was no proof in any case that an icterogenic agent was transferred by the transfusion the evidence being purely presumptive

between thirty six days and seven months and averaging between two and three months They showed that the occurrence of the jaundice was not directly due to an infection with the yellow fever virus and considered that there was presumptive evidence that the post inoculation jaundice was identical with infective hepatic jaundice Findlay MacCallum and Murgatroyd (58) later showed that the icterogenic agent bore no relationship to the strain of yellow fever virus used but that experiments suggested the presence of an extraneous virus in the human serum used in the preparation of the vaccine In 1939 Fox Manso Penna and Para (59) published an account of an outbreak of inoculation jaundice in Brazil after over a million people had been inoculated there against yellow fever Out of a group of 304 persons inoculated with one batch of vaccine 27% developed jaundice four to five months later one case proving fatal The occurrence between January 1st and July 1st in 1942 of 28 585 cases of jaundice 62 of which proved fatal out of between two and two and a half million American troops inoculated again t yellow fever was commented on in Editorials in the American medical press (60) Widespread attention was directed to this variety of jaundice which was again shown to be due to the presence of an icterogenic agent in the serum used as a vehicle in the preparation of the vaccine The latent period in this series varied between twelve and eighteen weeks In 1944 Findlay Martin and Mitchell (61) reported on a series of 689 cases of jaundice following yellow fever vaccine injections given during 1942 the average latent period being 101 5 days There was one fatal case Human serum is no longer used in the preparation of the vaccine and this type of jaundice has now ceased

Jaundice following Measles Convalescent Serum In the Annual Report of the Chief Medical Officer of Health for 1937 (62) there was a brief account of the occurrence of 37 cases of jaundice 7 of which proved fatal 16 to 100 days after the injection of measles convalescent serum from two small batches into between 82 and 109 recipients Propert (63) in 1938 drew attention to the cases of 7 children inoculated with convalescent measles serum of the same batch who all developed jaundice 78 to 83 days later 3 cases proving fatal The pooled serum used was the same as that referred to in the Ministry of Health report A fuller account was reconstructed from contemporaneous records and published in a Memorandum

orderlies at a venereal disease clinic who helped in the injections and had their hands contaminated with blood from the patients developed jaundice presumably through scratches in the skin Beattie and Marshall (72) tried to lower the incidence of post arsenical jaundice at a clinic by giving the patients a casein digest a casein digest with cystine or methionine These measures had no effect on the over all incidence of liver damage and jaundice but tended to shift its incidence to later in the course of treatment and to render the jaundice less severe

Jaundice following the Injection of Pentothal and Penicillin Darmady and Hardwick (74) observed 6 cases of jaundice following the intravenous injection of Pentothal between thirty one and one hundred and fifty one days previously and 4 cases sixty four to one hundred and thirteen days after the intramuscular injection of penicillin for the treatment of gonorrhœa or syphilis They suggested that here again the jaundice resulted from the use of dirty syringes They further considered that some cases of transfusion hepatitis may be due to the same cause and not to the blood injected Pentothal or penicillin having been given at the time of the transfusion Howells and Kerr (75) treated 47 patients suffering from jaundice who had previously received injections of penicillin the average latent period being 97 days Of these cases 41 had been given penicillin for the treatment of gonorrhœa or syphilis They concluded that penicillin is not the direct cause of the hepatitis which may be due to some icterogenic agent—*e g* a virus—transmitted by faulty injection technique

Jaundice following the Withdrawal of Blood Graham (76) was puzzled by the occurrence of 28 cases of jaundice developing during a period of two and a half years in patients suffering from diabetes mellitus treated at hospital none being noted in private patients during the same period He wrote I have come to the conclusion that the jaundice must be due to an infective agent passed from patient to patient at hospital He suspected that the infection was conveved by the syringes This might have occured when blood was removed for sugar estimations or when insulin was injected Droller (77) described an outbreak of jaundice in a diabetes clinic in which 62 patients were affected during a period of two years 4 of the cases proving fatal Blood was collected for sugar estimations in syringes which were not boiled

In a series of 760 recipients of transfusion of large pool plasma Lehane et al (70) found 54 cases (11 9%) in whom jaundice was a sequela in their opinion 0 35% of donors carry the icterogenic virus

We now pass on to the other type of serum jaundice in which it is believed that the causative agent is transmitted by contaminated syringes

Post-arsenical Jaundice Bigger (71) in 1943 drew attention to the increased incidence of post arsenical jaundice in venereal disease clinics In one of these the case incidence rose from 4 2% in 1941 to 16 5 % in 1942 Beattie and Marshall (72) found an incidence of over 50% of post arsenical jaundice at a clinic in 1942 and 1943 the jaundice usually developing between the ninety first and one hundred and sixty sixth day from the first injection Bigger (71) showed that the icterogenic agent was probably transmitted by syringes contaminated by a trace of blood from an infective patient and hence the term syringe jaundice' or 'syringe transmitted hepatitis came into use When an intravenous injection is given a minute amount of blood enters the syringe after the needle is put into the vein, and Bigger using in his experiments citrated blood contaminated with staphylococci showed that the methods in use at the clinics for sterilising the syringes such as soaking in 1/1 000 biniodide of mercury and washing in four changes of water were not adequate to remove minute quantities of infected material He recommended that the syringes should be boiled between each patient It was subsequently proposed by the Medical Research Council Syringe Sterilisation Committee that all glass syringes should be sterilised by dry heat at 160 C for an hour Salaman King Williams and Nicol (73) suggested that the increase in post arsenical jaundice during these years might be due to a variety of causes such as a shortage of syringes and an increase in the number of patients with a consequent diminution of the time the syringes could be soaked in disinfectants and also to an increased incidence of infective hepatitis if we accept the theory that the blood from patients incubating infective hepatitis is the source of the icterogenic agent There is a further possibility the low protein content of the war time diet or increased alcoholic consumption may have diminished the resistance of the liver to the icterogenic agent Sheehan (25) observed that some

Mitchell (61) in 1944 failed to produce jaundice in volunteers by the intranasal instillation of whole blood or the subcutaneous injection of whole blood from patients suffering from post inoculation jaundice MacCallum and Bauer (81) showed that serum from a presumed case of homologous serum jaundice was icterogenic when inoculated subcutaneously or intramuscularly into a volunteer on the seventh day after the onset of jaundice but not so fifty nine and one hundred and thirty four days later A further point in favour of the view that infective hepatitis and serum jaundice are manifestations of the same disease is afforded by the work of Dible McMichael and Sherlock (15) They examined micro scopically specimens obtained by liver puncture and could detect no difference in the appearance of the liver in infective hepatitis post arsenical jaundice and serum jaundice On the other hand there is evidence that infective hepatitis and serum jaundice are not due to the same virus Homologous serum jaundice does not afford immunity against infective hepatitis Sufferers from serum jaundice have not been proved to infect contacts although there is some evidence that this may very occasionally happen The incuba tion period of the two diseases is very different about thirty days for infective hepatitis and about one hundred days for serum jaundice Some authorities have also pointed out differences in the clinical features especially the occurrence of erythema multi forme stiff joints splenomegaly and the usually apyrexial course in serum jaundice MacCallum (82) has summed up the position as follows There are many debatable points in favour of and against the identity of infective hepatitis and homologous serum jaundice but there is no conclusive evidence one way or the other It is essential to find a satisfactory susceptible experi mental animal before much further knowledge can be obtained

Liver Puncture Biopsy

Historical One of the earliest recorded examples of diagnostic liver puncture is that performed by Stanley (83) in 1833 for drainage of a hepatic abscess In 1895 Lucatello (84) introduced the practice of exploratory puncture of the liver using a needle 1 mm in diameter The little fragments withdrawn were then lacerated and examined fresh but it was not possible to establish a histo pathological diagnosis of the disease process Von Hansemann (85) in 1904

and it was thought that small amounts of infective material might have contaminated the needles Damodaran and Hartfall (78) observed cases of jaundice in a clinic at which patients suffering from arthritis were treated with gold injections They do not state whether blood was removed for sedimentation readings In 1935 there were no cases of jaundice but later when there was a sparse outbreak of infective hepatitis up to 9 4% of the patients at the clinic developed jaundice They did not recognise infected syringes as a possible cause of the jaundice Sheehan (25) in the same year reported cases of jaundice occurring in a sanatorium It was noted that a patient who developed jaundice had had a vein puncture for the estimation of the sedimentation rate of the red cells about *three months earlier and on the same day as another patient in the* ward who was destined to develop jaundice a month or two later the incubation period being therefore about twelve weeks In all these cases it is is probable that the syringe was contaminated by a small quantity of blood containing the icterogenic agent and that as was shown by Mendelssohn and Witts (79) on removing the needle from the vein if the tourniquet is released before the needle is withdrawn a small quantity of blood is sucked back from the needle and the syringe into the vein If the syringe therefore is not sterile the patient is exposed to infection

The Nature of the Icterogenic Agent It has been suggested that infective hepatitis and homologous serum jaundice are both due to the same agent a virus but there is no proof of this view There is indeed evidence that the two conditions are dissimilar and result from different causes

The icterogenic agent has been found to behave in many respects like a virus it passes through Seitz filter pads and resists freezing and drying for long periods but it has never been transmitted to the lower animals Findlay and Martin (80) in 1943 instilled nasal washings from patients suffering from jaundice following yellow fever inoculation intranasally into volunteers In 3 out of 4 instances jaundice resulted at intervals of twenty eight thirty and fifty six days They suggested that infective hepatitis and homologous *serum* jaundice are due to the same virus the difference in the incubation period being due to the infection occurring intranasally in infective hepatitis and by subcutaneous injection in homologous serum jaundice Findlay Martin and

Mitchell (61) in 1944 failed to produce jaundice in volunteers by the intranasal instillation of whole blood or the subcutaneous injection of whole blood from patients suffering from post inoculation jaundice MacCallum and Bauer (81) showed that serum from a presumed case of homologous serum jaundice was icterogenic when inoculated subcutaneously or intramuscularly into a volunteer on the seventh day after the onset of jaundice but not so fifty nine and one hundred and thirty four days later A further point in favour of the view that infective hepatitis and serum jaundice are manifestations of the same disease is afforded by the work of *Dible McMichael and Sherlock (15) They examined micro* scopically specimens obtained by liver puncture and could detect no difference in the appearance of the liver in infective hepatitis post arsenical jaundice and serum jaundice On the other hand there is evidence that infective hepatitis and serum jaundice are not due to the same virus Homologous serum jaundice does not afford immunity against infective hepatitis Sufferers from serum jaundice have not been proved to infect contacts although there is some evidence that this may very occasionally happen The incubation period of the two diseases is very different about thirty days for infective hepatitis and about one hundred days for serum jaundice Some authorities have also pointed out differences in the clinical features especially the occurrence of erythema multiforme stiff joints splenomegaly and the usually apyrexial course in serum jaundice MacCallum (82) has summed up the position as follows There are many debatable points in favour of and against the identity of infective hepatitis and homologous serum jaundice but there is no conclusive evidence one way or the other It is essential to find a satisfactory susceptible experimental animal before much further knowledge can be obtained

Liver Puncture Biopsy

Historical One of the earliest recorded examples of diagnostic liver puncture is that performed by Stanley (83) in 1833 for drainage of a hepatic abscess In 1895 Lucatello (84) introduced the practice of exploratory puncture of the liver using a needle 1 mm in diameter The little fragments withdrawn were then lacerated and examined fresh but it was not possible to establish a histo pathological diagnosis of the disease process Von Hansemann (85) in 1904

used a special trocar to obtain small fragments of liver tissue and by this method made a diagnosis of syphilis of the liver In 1927 Shupfer (86) employed a special bevelled needle with an internal diameter of 1 4 mm attached to a 10 ml syringe By this means he obtained specimens of liver up to 2 cm in length which were usually sufficient for a histological diagnosis Iversen and Roholm (87) in 1939 introduced their method of liver puncture with a cannula of 2 mm bore through which they aspirated specimens of liver tissue suitable for paraffin sections and histological examination This has lead to a widespread use of aspiration liver biopsy as a reliable aid to diagnosis

The Indications Aspiration or as it is sometimes called needle liver biopsy is of value in enabling the nature of pathological changes in the liver to be determined during life without the necessity of a laparotomy By this procedure many conditions affecting the liver may be diagnosed amongst which may be mentioned infective hepatitis obstructive jaundice cirrhosis neoplasms fatty changes amyloidosis sarcoidosis leukæmia Gaucher s disease reticuloses miliary tuberculosis Kala azar schistosomiasis hæmochromatosis infectious mononucleosis and brucellosis

The Contra indications Because of the risk of spreading infection it is wise not to perform liver puncture in suspected cases of liver abscess or hydatid disease or if there is infection in the lungs or pleura It is contra indicated if there is severe anæmia or a tendency to bleeding which does not respond to treatment It is also best avoided when the liver dulness cannot be delineated by percussion owing to emphysema or in ascites unless the fluid has been previously removed It should never be attempted if the patient is uncooperative and unwilling or unable to hold his breath Some authorities such as Cogswell et al (88) consider it unwise to perform liver biopsy in passive congestion due to heart failure owing to the possibly increased risk of bleeding

The Method The prothrombin bleeding and clotting times and the blood group of the patient should be determined and two pints of compatible blood should be at hand with facilities for transfusion in case of hæmorrhage following the puncture Twenty mg of Synkavit are injected intramuscularly daily for three days or longer if necessary before the biopsy is performed The puncture should

not be made until or unless the prothrombin bleeding and clotting times are normal If there is ascites the fluid should be aspirated so that the liver comes in close apposition to the chest wall It is usually recommended that morphine sulphate gr 1/6 to 1/4 should be injected subcutaneously half an hour before the operation but in cases of advanced cirrhosis such a dose of morphine may prove dangerous or even fatal and should not be given In such instances sodium phenobarbitone gr 1 in 1 ml of water may be injected intramuscularly half an hour before the puncture The patient lies in bed on his back with his right hand behind his head the side of the chest parallel with the edge of the bed and the left buttock raised on a pillow so as to tilt the body slightly to the right The upper limit of the hepatic dulness is percussed out with the breath held in full expiration and the puncture site selected one intercostal space below the upper border of the liver dulness just behind the anterior axillary line Thus is usually the seventh intercostal space The skin over the proposed site of puncture is painted with iodine and the skin subcutaneous tissues intercostal muscles pleura diaphragm and peritoneum down to the capsule of the liver are freely infiltrated with at least 10 ml of 2 % procaine hydrochloride solution The patient should now be instructed how to breathe in breathe out deeply and then stop breathing It is explained that he will be told to do this when the puncture is being made and that he must hold his breath in full expiration until the word is given that he may breathe again

For the puncture a trocar with a handle and cannula are used The cannula is 15 cm long with a bore of 1 8 mm and its end is bevelled and sharp It is fitted with a metal guard and can be attached by an adaptor to a 20 ml all glass syringe The skin at the site of the puncture is nicked with a tenotomy knife and the sterilised trocar and cannula are pushed slowly through the intercostal space until the transmitted respiratory movements indicate that the diaphragm has been reached The guard is now adjusted to allow the cannula to be pushed 3 cm into the liver the trocar is withdrawn and the syringe is fitted to the cannula The patient is now told to breathe in breathe out fully and stop breathing It is essential for him to realise that he must hold his breath until told he may breathe again The cannula is now inserted sharply and with a slight twisting movement into the liver until the guard is in contact

with the skin The syringe plunger is withdrawn and the cannula and syringe removed maintaining suction as this is done The patient is now told he may breathe again The cannula is only in the liver for about five seconds The biopsy specimen is found either in the cannula or in the syringe and is placed in a specimen *bottle containing 10% formol in saline* The wound is sealed with collodion and the patient instructed to lie flat for twenty four hours The pulse rate should be taken every fifteen minutes for two hours and every hour for twenty two hours and if it rises the physician must be sent for If there are signs of internal hæmorrhage a blood transfusion is given There may be pain on the right side of the chest or at the top of the right shoulder requiring an analgesic such as aspirin or Veganin

Variations in Technique A cannula of 2 mm bore with three sharp teeth was used by Iversen and Roholm (87) but this may increase the risk of tearing the liver capsule with subsequent hæmorrhage Sherlock (89) uses a trocar and cannula with a bore of 1 mm and inserts the cannula into the liver for ½ inch before removing the trocar and then pushes the cannula into the liver a further 4 to 5 cm Silverman (90) in 1938 introduced a split needle for obtaining liver specimens It consists of an outer needle or cannula of 16 gauge and an inner needle of 18 gauge which is split longitudinally down to the hub The outer needle or cannula with obturator in place is inserted into the liver The obturator is then removed and the split needle inserted in its place and pushed into the liver cutting a cylinder of tissue between its prongs The cannula is now advanced over the split needle into the liver for a distance of about ½ inch The entire instrument is then rotated slightly and the inner needle withdrawn with the specimen A Vim Silverman needle was employed by Cogswell et al (88) the fragment of liver tissue removed being 1–2 mm in diameter and 1–2 cm long Terry (91) used a modified Gillman needle in which the stylet is attached to the plunger of the syringe The needle has a diameter of 1 8 mm with a bevelled point internally bored to give a knife like edge Other sites of puncture are sometimes preferred Thus Iversen and Roholm (87) punctured the ninth intercostal space in the posterior axillary line Gillman and Gillman (92) the costo xiphoid angle between the xiphoid and the right costal margin and Davis et al (93) used the subcostal route The needle either a

Fig 7 Liver biopsy showing fatty changes

Fig 8 Liver biopsy showing secondary carcinoma

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Silverman or a Roth Turkel needle with small saw teeth was inserted a few centimetres to the right of the xiphoid process and below the costal margin and directed upwards and to the right toward the centre of the liver

Some Difficulties and Complications Failure to obtain a satisfactory specimen of liver may be due to the presence of ascites increasing the mobility of the liver The chief difficulty in detaching and removing a suitable piece of liver is met with in cirrhosis The risk of fatal hæmorrhage has diminished with improvements in technique and with the use of vitamin K Terry (91) refers to 2 469 biopsies performed between 1939 and 1948 in which there were 7 deaths Cogswell et al (88) performed 403 biopsies on 345 patients without any serious reaction Other complications include pneumothorax hæmothorax empyema and air embolism Davis et al (93) performed biopsies by the subcostal route on 68 patients without serious complications but state that the risks include extensive bleeding into the peritoneal cavity and abdominal wall perforation of the large or small intestine or gall bladder introduction of infection or spread of localised infection into the general peritoneal cavity and injury to the kidney adrenal or pancreas

Conclusions Liver biopsy is usually but not invariably not dangerous or painful when performed by a skilled physician and when due precautions are taken but it should not be undertaken sporadically by the uninitiated We have found it of value in distinguishing between obstructive jaundice of short duration and jaundice due to hepatitis In the former the biopsy reveals accumulation of the bile in the liver canaliculi near the centre of the lobules in the latter there is loss of lobular structure ballooning of the polygonal cells and focal areas of small mononuclear cell infiltration The early fatty changes in cirrhosis are well shown (see Fig 7) and enable a diagnosis to be made when the changes are still reversible with adequate dietetic treatment We have also found the method of value in the diagnosis of secondary carcinoma of the liver (see Fig 8) thus avoiding an unnecessary laparotomy At times also a correct diagnosis may be made in obscure cases of hepatomegaly

Liver Function Tests

Investigation of the Pigmentary Function of the Liver Hæmoglobin released from erythrocytes destroyed in the reticulo

endothelial system is converted to bilirubin and biliverdin which are excreted in the bile The probable route is (i) the porphyrin ring is opened leading to the formation of a verdohæmoglobin — probably a biliverdin—globin iron complex Lemberg et al (94) (ii) the iron is split off to travel thereafter with the β globulin fraction of the plasma proteins and some of the (green) biliverdin is reduced to (orange) bilirubin which remains in association with globin

Authorities disagree regarding the nature of the protein binding of the plasma bilirubin and regarding changes taking place in this linkage when the bilirubin protein complex passes through the liver (Watson (95)) The subject is critically discussed by With (96) The consensus of present day opinion is that the pre hepatic (type A indirect type I) bilirubin is closely bound to protein it is converted to post hepatic (type B direct reading type II) bilirubin probably in the Kuppfer cells and is excreted by the parenchymal cells (Schaffner et al (97)) This bilirubin is only loosely associated with protein and is probably in the form of a sodium salt

In the intestines bilirubin is reduced by bacterial action to *urobilinogen* (a mixture of two colourless chromogens *mesobili rubinogen* and stercobilinogen) Some of the urobilinogen is excreted in the fæces and is oxidised by the air to urobilin the remainder is absorbed into the portal circulation and returned to the liver where a part is excreted unchanged a part re oxidised and excreted as bilirubin and a part returns to the general circulation and is excreted by the kidney

Types of Jaundice and the van den Bergh Reaction

In view of the easily available literature (*e g* McNee (98)) only the briefest summary of this important work will be given here Jaundice at the time of Virchow was considered to fall under *two* headings hepatogenous and anhepatogenous Thus in the hepato genous variety the condition resulted from obstruction of the biliary tract whilst in the second anhepatogenous variety the bile pig ment was thought to originate in tissues other than the liver Later owing to certain experimental work which need not be detailed here the possibility of anhepatogenous jaundice was excluded Thus Eppinger (99) stated that all jaundice was obstructive and in those cases where obvious obstruction could not be seen the seat

of the trouble was said to be in the fine bile capillaries Later however the work of Aschoff (100) and his co-workers pointed rather to the original separation of jaundice into hepatogenous or obstructive and hæmolytic or anhepatogenous His work has completely changed the views on the formation of bile pigments from hæmoglobin Thus it used to be thought that this change was effected by the polygonal cells of the liver but in the newer conception it is held that these simply pass the pigment on from another group of cells to the lumen of the bile capillary

Jaundice may be classified either according to the site of the primary disturbance into obstructive post hepatic hepatic or pre hepatic (McNee (98)) or alternatively depending on the path taken by the excess bilirubin into retention jaundice if the bile has not passed through the liver cells and regurgitation jaundice if this has taken place A combined classification is shown in the table after Lichtman (101)

SITE	CAUSE	TYPE OF JAUNDICE
Pre hepatic	Excessive destruction of R.B C.s	Retention
Hepatic		
Intralobular	(i) familial hyperbilirubinæmia Meulengracht (102) due to subnormal hepato-cellular function	Retention
	(ii) parenchymal cell necrosis or œdema (toxic or infective hepatitis)	Regurgitation
	(iii) cholangiolitis	Regurgitation
Extralobular	(iv) cholangitis	Regurgitation
	(v) distortion of cell-canalicular continuity (as in cirrhosis)	Regurgitation
Post hepatic	Obstruction to biliary outflow (stone carcinoma etc)	Regurgitation

In retention jaundice the excess bilirubin is principally type A in regurgitation jaundice type B A chemical reaction to distinguish between these would have diagnostic value

This has been devised by Hijmans van den Bergh (103) who employs Ehrlich s azo reaction This consists in adding to the solution of bilirubin a diazonium salt in acid solution causing the appearance of a purple compound azo-bilirubin The diazonium compound is provided by mixing a solution of sulphanilic acid in HCl with sodium nitrite Van den Bergh noted that icteric sera, when treated in this manner either gave an immediate colour or else a very delayed reaction He was able to assert that the first type of reaction was given by bilirubin which had passed through the liver cells

and that the delayed reaction resulted with pigment formed without the agency of the polygonal cells i e absorbed direct from the Kupffer cells The present day view is that colour developing within a minute of adding the diazo reagent to the serum (prompt direct reaction) is probably a measure of bilirubin type B colour developing when the mixure is allowed to stand or on adding alcohol (Ducci and Watson (104)) or on adding caffeine and urea (Gray and Widbourne (105)) is a measure of total bilirubin (A + B)

Interpretation of the Results Normal serum contains both bilirubin A and B approximately 30% being type B In clinical work the only significant observations are of total serum bilirubin (as measured by any suitable method) and of the presence of a prompt direct reaction a visible colour appearing within a minute of adding the diazo reagents to the serum 'Biphasic and delayed direct reactions have no significance Many workers such as Klatski and Drill (106) consider that a positive direct reaction or alteration of the prompt total serum bilirubin ratio is of no diagnostic significance whilst contrariwise Schaffner et al (97) find that the average percentage of bilirubin B is significantly higher in obstructive jaundice and toxic hepatitis than in infective hepatitis In our experience a raised total serum bilirubin with a negative prompt direct reaction indicates retention jaundice if the prompt direct reaction is positive (more than 45% being type B) then there is *regurgitation* jaundice The total serum bilirubin level serves to follow the progress of the disease as clinical jaundice may not be noticeable for several days after the serum level is raised and the pigment will remain in the skin for a little while after the serum level has returned to normal Jaundice generally appears when the serum bilirubin is in the range 1 5–2 mg % In retention jaundice the range is 0 5–1 mg % higher for bilirubin type A

Icterus Index As this measures only the colour of the serum and not chemically its bilirubin content other materials (*e g* carotenoids hæmatin) are also detected and the test is of little value except as a simple method for the small laboratory to follow the degree of jaundice It may have value in the control of biliverdin jaundice (Larson et al (107)) in cases of long standing obstruction or of receding jaundice for biliverdin does not give a *Van den* Bergh reaction

Biliary Pigments in the Urine The kidney does not excrete

bilirubin type A (hence the term acholuric jaundice) whilst bile pigments can be detected in the urine by simple tests when the total serum bilirubin level in cases of regurgitation jaundice is more than approximately 2 mg / This threshold is not a true one and in hepatitis urinary bile pigments can be found at serum bilirubin levels of about 1 mg /

Normally a small quantity of urobilinogen is detectable in freshly passed urine by the use of Ehrlich s aldehyde reagent In retention jaundice (due to overproduction of bilirubin) the urine and fæcal urobilinogen are greatly increased in the obstructive type of regurgitation jaundice urobilinogen is absent from both urine and fæces In early liver damage *e g* in infective hepatitis one of the first hepatic functions to be altered is the power to oxidise urobilinogen to bilirubin hence an increased urinary urobilinogen is a sensitive sign of minimal impairment As the liver damage proceeds an obstructive element supervenes and the urinary urobilinogen disappears to return on recovery Watson (108) discusses urobilinogen excretion in greater detail and gives quantitative details of the findings in various diseases

The Bilirubin Excretion Test The measurement of the excretion of bilirubin injected intravenously as a means of estimating hepatic function was originated by Eilbott (109) The technique described below is that of Soffer and Paulson (110) The test is however rarely used today

A total amount of bilirubin equal to 1 mg per kg of body weight but not exceeding 70 mg is dissolved in 15 ml M/10 sodium carbonate which previously has been boiled and then cooled to 80 C A control sample of blood is collected in a dry syringe to prevent hæmolysis after which with the needle *in situ* the bilirubin solution is injected Further specimens of blood are taken five minutes later and after four hours Two ml of plasma from the control and from the four hour specimens are treated with 2 ml of acetone and centrifuged while 1 ml of the plasma from the five minute specimen is treated with 4 ml of acetone The supernatant fluids are filtered and promptly matched against suitable standards These are prepared from stock 1 in 2 000 solution of potassium dichromate which yields a colour identical with that of 1 mg of circulating bilirubin per 100 ml plasma Suitable standards contain the equivalent of 0 9 0 8 0 7 0 6 0 5 0 4 0 3 and 0 2 mg of circulating bilirubin

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Biliary Pigments in the Urine The kidney does not excrete

Findings Mateer et al (115 and 116) using this test compared it with the oral method and found that the intravenous test is more delicate yielding positive results (diminished hippuric acid excretion) in 85% more cases of liver damage than does the oral test The test is sensitive but has the disadvantages that the degree of positivity is not proportional to the extent of liver damage and that false low outputs of hippuric acid are found if there is renal damage dehydration or obstructive anuria The test is applicable to jaundiced and non jaundiced patients it is not very reliable if used in the differential diagnosis of hepatic from post hepatic jaundice as minor degrees of obstruction generally give a positive result

Investigation of the Capacity of the Liver to Excrete Foreign Substances

The Bromsulphthalein Test Rosenthal and White in 1924 (117 and 118) proposed a liver function test based on the ability of the liver to remove bromsulphthalein (phenoltetrabromphthalein disodium sulphonate) from the blood The dye in the plasma is bound loosely to albumin and the rate of removal from the plasma by the liver is a constant proportion of the plasma level and depends on the blood flow to the liver and the functional capacity of the parenchymal cells Ingelfinger et al (119) discuss variation in this percentage disappearance rate normally 10–16%/min In very severe liver damage there is some extra hepatic bromsulphthalein uptake (Mendeloff et al (120))

For clinical purposes a simple satisfactory method of estimating bromsulphthalein clearance is as follows The test is performed with the patient fasting and at rest Inject intravenously bromsulphthalein solution 5 mg/kg body weight Some authorities use a 2 mg/kg dose this does not give a sufficient load to the parenchymal cells and we have not observed any of the reported toxic effects of the higher dose At five minutes and at forty five minutes after the injection withdraw 10 ml samples of blood for estimation of the dye content of the sera

Analysis of the five minute sample serves as a check on the correctness of the quantity injected normally it contains between 15% and 85% (compared with the 100% standard) At forty five

per 100 ml of plasma and are made by diluting 9 8 7 6 5 4 3 and 2 ml of the 1 in 2 000 dichromate to 10 ml with water When acidified with one drop of dilute hydrochloric acid these standards *are stable for some time and may be kept in sealed tubes*

Calculation 200 × (mg % bilirubin found in four hour filtrate — mg bilirubin found in control filtrate) divided by (5 × mg % bilirubin found in five min filtrate — 2 × mg % bilirubin found in control filtrate) gives the percentage of bilirubin retained

Findings Normally not more than 5 to 6% of the injected bilirubin should be retained at the end of four hours An advantage of the test is that the liver is given a natural pigment bilirubin to excrete although this substance is expensive and difficult to procure It should be borne in mind that the test is inapplicable to patients with jaundice and is most useful in cases in which the bilirubin does not exceed 1 mg per 100 ml plasma Several workers have reported favourably on the reliability and delicacy of the bilirubin test Thus Soffer (111) found that the normal retention of bilirubin was exceeded in recovering cases of diffuse hepatitis in which the jaundice had subsided as well as in cases of cirrhosis and malignancy Sweet Gray and Allen (112) concluded that the test was more delicate than the galactose tolerance brom sulphthalein and hippuric acid tests in the detection of hepatic damage in cases of minimal or no symptomatic evidence of cirrhosis of the liver

Investigation of the Antitoxic Powers of the Liver

The Oral Hippuric Acid Excretion Test This test which was devised by Quick (113) depends on the ability of the liver to conjugate benzoic acid with glycine It has now been largely replaced by the intravenous test

The Intravenous Hippuric Acid Test In 1938 Quick et al (114) showed that hippuric acid is synthesised in man following the intravenous injection of sodium benzoate 1 77 g of sodium benzoate is dissolved in 20 ml of distilled water and injected slowly intravenously taking five minutes over the injection The bladder is emptied before the test and the urine is collected one hour after the injection The hippuric acid is estimated in the urine Normally 0 7 g to 0 95 g is excreted as hippuric acid in the first hour

hepatitis damage to the liver cells results in changes in the plasma proteins detectable by the normal methods of quantitative analysis the albumin level being decreased and the globulins increased In acute parenchymal hepatitis (*e g* infective hepatitis) the changes in the proteins are not detectable by methods of routine analysis there is a qualitative alteration in the albumin and an increase of the γ globulin fraction

In order to detect these qualitative alterations of the plasma proteins tests are used in which the patient s serum is added to a suitable colloidal system condition are arranged so that normal (but generally not hepatitic) albumin stabilises the system and inhibits flocculation whereas generally speaking γ globulin in excess (especially hepatitic globulin) destroys the stability and flocculation or precipitation results (MacLagan (124)) A very large number of such tests have been developed depending on the nature of the precipitant and the conditions of the test amongst those in use are tests employing zinc sulphate (Kunkel (125)) cadmium sulphate (Wunderly and Wuhrmann (126)) and colloidal red (Ducci (127))

The tests that we employ are as follows —

Thymol Turbidity (MacLagan (128)) The reagent used is a buffered solution of thymol the resultant turbidity is read in comparison with artificial standards (Normal 0–4 units) Precipitation occurs with excess β and γ globulins and with a lipoid factor and is inhibited by normal albumin

Colloidal Gold (MacLagan (128)) The reagent used is a colloidal gold solution as employed for the Lange test on cerebro spinal fluid When the test is done using serum instead of a colour change precipitation occurs and degrees of precipitation 0–5 are distinguished (Normal 0–1) Precipitation occurs with excess γ globulin and is inhibited by normal α and β globulins and by albumins only slightly

Cephalin Cholesterol (Hanger (129) Steinberg (130)) The reagent now used in our laboratories is an artificial emulsion of desoxycholic acid + cholesterol replacing the original emulsion of brain extract + cholesterol Complete precipitation is termed 4 + (normal 0–1 +) Precipitation occurs with excess γ globulin and hepatitic α and β globulins and is inhibited by albumin

These tests in general give parallel results and are much used in

minutes the dye content of the serum is normally between 0 and 5% a value above 10% indicates definite liver damage and the height of the forty five minute level is proportional to the degree of damage

This test is sensitive and is most satisfactory for the detection and investigation of diffuse hepatocellular damage in the absence of jaundice it should be noted that in children bromsulphthalein excretion does not follow that of bilirubin The test cannot be used to differentiate hepatic disease and post hepatic obstruction (which equally causes retention of dye) mildly positive results are often also found in heart failure and febrile states At the time of writing the dye was obtainable only from the U S A and the cost of material per injection was about 15s

INVESTIGATION OF THE METABOLIC FUNCTIONS OF THE LIVER

Carbohydrate Of the three sugars that could be used in tests of tolerance glucose and lævulose are not satisfactory for liver function tests because of their extra hepatic utilisation MacLagan (121 and 122) has used galactose with consistent results

Galactose Tolerance Test This sugar is absorbed freely from the gut and readily stored as glycogen by the normal liver More satisfactory results are obtained if the patient is fasting Take a resting blood sample the patient then empties the bladder completely and drinks a solution of 40 g galactose in lemon water blood samples are taken every thirty minutes for two hours and the total urine excretion collected over five hours Normally the blood galactose level reaches a peak at sixty minutes of 10–40 mg /100 ml the sum of the 4 blood galactose values—termed the galactose index —is normally about 70 mg an index over 160 indicates hepatocellular damage Normally the urinary excretion in five hours is less than 3 g an excretion of over 4 g must be regarded with suspicion an excretion of over 6 g indicates severe hepatocellular damage

This test is moderately sensitive and has the advantage of giving normal results in most cases of uncomplicated mechanical obstructive jaundice (Bensley (123)) The test gives invalid results if there is malabsorption of the carbohydrate from the gut as in the sprue syndrome or myxœdema

Protein The plasma proteins are made in the liver In chronic

the back pressure of the pent up biliary secretions to produce excess of the enzyme

The methods of assay generally in use in this country (modifications from King and Armstrong (133)) involve incubation of the serum with a buffered phenyl phosphate solution then estimating the phenol liberated under standard conditions—the normal being 3–13 King Armstrong units in adults In the U S A the method of Bodansky (134) is commonly employed involving the measurements of phosphorus liberated from glycerophosphate the normal being 1–4 units in adults

This estimation is valuable in the differential diagnosis of obstructive from hepatocellular jaundice especially when used in combination with the simple flocculation tests In an obstructive jaundice the level is generally above 35 units (King Armstrong) whilst in hepatocellular jaundice it is usually between 10 and 25 units High values are also found in cirrhosis of the liver more especially in biliary cirrhosis and sometimes in widespread carcinomatosis It must be remembered in interpreting alkaline phosphatase levels that the enzyme is also produced by osteoblasts and high values are obtained in such bone diseases as rickets hyperparathyroidism primary and secondary malignant disease and Paget s disease

The Prothrombin Time Prothrombin is synthesised in the parenchymal cells of the liver The production depends on the amount of vitamin K reaching the cells and on the functional state of the cells Accordingly in liver disease the production of prothrombin may be diminished for two reasons (i) damage to parenchymal cells (ii) absence of bile salts from the intestine leading to diminished absorption of vitamin K

Estimation of the prothrombin time by itself is not satisfactory as a liver function test To differentiate between low prothrombin times resulting from liver damage and from deficient absorption of vitamin K recourse may be had to the intramuscular injection of a suitable preparation of vitamin K (*e g* 2 mg of 2 methyl 1 4 naphthaquinone Lord and Andrus (135)) this returns the prothrombin time to normal when deficiency is caused by malabsorption but not when caused by hepatocellular damage

The Serum Cholinesterase This enzyme which destroys acetylcholine is a protein synthesised by the liver and attention has been paid in recent years to its estimation as an index of liver

the differential diagnosis of hepatic from post hepatic jaundice In hepatic disease the tests give positive results in an uncomplicated obstruction the tests tend to be normal

When used in the investigation of liver function the cephalin cholesterol test is the most sensitive and best employed in the detection of liver damage in the absence of jaundice It is unsuitable for differential diagnosis as it frequently yields a positive result in apparently uncomplicated obstructive jaundice If the thymol or gold test is positive in obstructive jaundice then there is super imposed liver damage The flocculation tests are frequently negative in carcinomatosis or compensated cirrhosis of the liver

The thymol turbidity test is often positive in diseases associated with hyperlipæmia in the absence of any protein changes and has been sometimes found to give positive results in glandular fever The colloidal gold test tends especially to yield 'false positives' in rheumatoid arthritis and the cephalin cholesterol in rheumatic heart disease and malaria though all these tests are sensitive to such miscellaneous diseases (with possibly some secondary liver damage) associated with changes in the globulins

Lipoid Alterations in the serum levels of free and ester cholesterol occur in various types of liver disease (Albrinke et al (131)) however these changes are not generally employed for diagnostic purposes

Obstructive jaundice	free cholesterol greatly increased ester cholesterol increased
Hepatocellular damage	free cholesterol variable ester cholesterol greatly diminished

In jaundice due to chemical agents (e g NAB) and during recovery from acute parenchymal damage an hypercholesterolæmia is frequently found

Investigations on Enzymes

The Plasma Alkaline Phosphatase Roberts (132) was the first to note an increase in the plasma alkaline phosphatase in some cases of obstructive jaundice The reason for this alteration is not known for certain The main hypotheses are that during obstruction the enzyme normally excreted in the bile is forced back into the general circulation that the parenchymal cells fail to excrete the enzyme, or that the parenchymal cells are 'stimulated' by

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function (McArdle (136)) The serum cholinesterase level is decreased in hepatocellular jaundice and is normal in uncomplicated obstructive jaundice The estimation is technically difficult, and the units of measurement depend on the method employed Vorhaus et al (137) describe studies using a simplified technique

Conclusions The choice of liver function tests to be performed on a patient depends on the purpose for which the tests are to be done As an aid to the differential diagnosis of jaundice the urine and fæces should be tested for bile and urobilinogen the Van den Bergh reaction and total serum bilirubin estimated and the combination of analyses for serum alkaline phosphatase and a flocculation test (*e g* colloidal gold) considered For the estimation of liver insufficiency the bromsulphthalein clearance the estimation of total and differential serum proteins and a flocculation test (*e g* cephalin cholesterol) are suitable

There is no single comprehensive test for liver function and the tests described each deal with limited functions of the liver Extension of abnormality detected in one aspect of liver metabolism to the conclusion that there is global damage to the liver is not always justifiable It must be emphasised that only when considered in conjunction with the more important clinical evidence can reliable conclusions be drawn from the results of liver function tests

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A biopsy attachment to the Wolf Schindler instrument has been developed in America The patient should be examined fasting using 2% Anethaine as a local anæsthetic A preliminary sub cutaneous injection of morphin sulph gr ¼ and atropin sulph gr 1/120 is given three quarters of an hour before instrumentation and a gr 1½ Anethaine pastille is sucked thirty minutes later If the pharynx is still sensitive 3 ml of 2% Anethaine are used in addition as a gargle Overdosage may result in toxic symptoms Rodgers (4) gives the following indications for gastroscopy When the skiagram has shown some abnormality but no definite diagnosis has been made or when a diagnosis having been made some additional information is required It is helpful in case of dyspepsia in which the skiagram has revealed no abnormality In certain cases of anæmia its use is of interest although of little practical value There are also contra indications which include local conditions such as trismus œsophageal stenosis enlarged mediastinal glands an aneurysm or severe curvature of the spine A large carcinoma near the cardiac orifice may render the examination impossible and it should not be performed within a few days of a severe hæmatemesis The abolition of the blind area in the stomach has been largely effected by the use of the Hermon Taylor gastroscope In some cases of doubtful X ray diagnosis of carcinoma a definite diagnosis either positive or negative may be made by gastroscopy On the other hand a lesion which has been seen through the gastroscope and diagnosed as an ulcer may be proved by operation to be a carcinoma some weeks later Like most methods of investigation gastroscopy is not infallible Gastric ulcers may frequently be visualised and be seen to heal under medical treatment In some cases an ulcer may be seen which is not demonstrable radiologically and the converse is also true Lintott (5) found that gastroscopic healing of gastric ulcers did not occur until one to several weeks after the disappearance of all X ray deformity A gastro jejunal stoma is not always easy to see

It is in the diagnosis of gastritis that gastroscopy appears most useful as has been emphasised by Bank and Renshaw (6) and by Freeman (7) During the active stages of inflammation the mucous membrane shows hyperæmia œdema exudation hæmor rhages and superficial ulceration The results of inflammation

CHAPTER VII

THE STOMACH

X ray Examination

Examination of the stomach and of the duodenum by means of the X rays after the ingestion of an opaque meal is a useful adjunct in the differential diagnosis of ulcers of these organs

Investigations should be made both under the screen and with films The opaque meal used in the screen examination usually consists of 3 oz of barium sulphate in half a pint of milk By this means the shape of the stomach and its filling properties can be investigated

The motor power and rate of stomach emptying are better studied with a more solid meal such as porridge or bread and milk containing barium sulphate There should be no residue of such a meal in the stomach after six hours

Persistent deformity in the outline of the stomach or duodenum affords some radiological indication of ulcer Thus in gastric ulcer the barium may be seen filling a niche in the wall of the stomach or there may be a definite organic hour glass appearance In the duodenum a persistent alteration in the shape of the duodenal cap may be seen The X ray examination will also reveal pyloric obstruction

Variations in tone or in the position of the stomach are not diagnostic of ulceration Subsequent examination after the administration of belladonna is of value in distinguishing between constriction of the stomach due to spasm and that due to cicatrisation

Study of the relief pattern of the gastric and duodenal mucous membrane has been stimulated by the work of Berg (1) Cordiner and Calthrop (2) have published details of this method as applied to the radiography of the duodenal cap and they emphasise its importance

Gastroscopy

The introduction of the flexible gastroscope in 1932 by Schindler and Wolf and of the Hermon Taylor (3) model with controllable flexibility in 1941 has further advanced the study of gastric diseases

records a mortality of 1 3 % in 368 cases of the hospital class treated by early feeding The more recent mortality figures are not so good Thus Avery Jones (14) in 1947 recorded a mortality rate of 8 % in a series of 687 cases admitted to hospital between 1940 and 1947 Baker (15) in a series of 576 cases admitted to Selly Oak Hospital between 1940 and 1945 found 13 4% proved fatal and Lewin and Truelove (16) in 1949 showed a mortality figure of 19 % in a series of 305 cases admitted as emergencies Needham and McConachie (17) have analysed the records of 476 cases of hæmatemesis admitted as emergencies to the Aberdeen Royal Infirmary from January 1941 to December 1948 The mortality rate was 13 9 %

Discussions at the Royal Society of Medicine London in 1924 and again in 1934 (18) emphasised that the immediate treatment of acute hæmatemesis due to gastric or duodenal ulcer is almost always medical It has usually been considered that the likelihood of death rises with the recurrence of hæmorrhage a primary hæmorrhage not being so dangerous as a subsequent one This view is not borne out by statistics from Seattle (U S A) Thus Blackford and Allen (19) in 1942 recorded 120 deaths in men and 31 deaths in women from bleeding peptic ulcer Seventy seven per cent of all fatalities occurred during the first hæmorrhage and only 8 deaths occurred in patients under forty five years old Needham and McConachie s (17) figures do not support this view They found the risk of dying was eight to ten times greater in the recurrent group In patients under forty years of age fatalities are almost unknown but the mortality rate rise steeply after middle age The surgical view point will be referred to later

Medical Treatment of Severe Hæmatemesis It must be clearly understood that the type of patient for whom this treatment is required is so desperately ill that he is not able to take feeds by mouth (*a*) The patient is put to bed kept warm and very still (*b*) Morphine sulphate gr $\frac{1}{4}$ and atropine sulphate gr $\frac{1}{100}$ are injected subcutaneously This may be repeated up to a total of morphine gr 1 in twenty four hours Willcox (20) has drawn attention to the danger of morphine in case of hæmatemesis due to cirrhosis of the liver when the detoxicating power of the liver may be so diminished that even gr $\frac{1}{4}$ may cause death our experience supports this view (*c*) Nothing is given by mouth for

are granularity of the mucous membrane, narrowing and distortion of the folds and atrophy Gill et al (8) examining 217 service cases of dyspepsia gastroscopically found gastritis as an isolated lesion in 32% and gastritis was present as an associated lesion in 80% of cases of gastric ulcer and in 60% of cases of pyloric and duodenal ulcer Berry (9) studied gastroscopically 100 alcohol addicts and found that only 35% had unequivocal alcoholic gastritis

In pernicious and simple achlorhydric anæmias the mucous membrane is thin pale and as described by Hartfall (10) the most characteristic feature in pernicious anæmia is the presence of branching submucosal vessels which impart a bluish network to the thin pale wash leather background The pallor is probably due to anæmia and is much less marked after adequate treatment but the mucous membrane remains thin and does not appear to regenerate The changes in the simple achlorhydric anæmias are very similar but the atrophy is less severe There does not appear to be any evidence of a preceding gastritis

Principles of Treatment of Gastric and Duodenal Ulcers

Varous modifications in the treatment of ulcers of the stomach and duodenum have been introduced during recent years From the practical point of view the subject can be considered under the following headings —

The treatment of hæmatemesis due to gastric or duodenal ulceration

The medical treatment of ulcers not at the time complicated by hæmorrhage

The indications for surgical treatment of gastric and duodenal ulcers not complicated by hæmorrhage

Hæmatemesis It is probable that between 10% and 30% of all peptic ulcers bleed but the hæmorrhage is not necessarily severe and the average death rate from hæmorrhage has been variously estimated as follows Cullinan and Price (11) give a mortality of 10% in a series of 105 cases of the hospital class 4 of which were treated surgically all dying Hurst and Ryle (12) in 67 cases of the private practice class had a mortality of 1 5% Three of their cases were treated surgically and all died Meulengracht (13)

alternately before three other feeds A teaspoonful of an alkaline powder (see p 163) or of Aludrox should be given after five feeds and a double dose at night Ferri et ammon citrat gr 30 should be given three times a day between the feeds The subsequent medical treatment is as for the treatment of ulcer not complicated by hæmorrhage and is detailed later

Routine Medical Treatment of Hæmatemesis In the majority of cases of hæmatemesis the patient can tolerate early feeding and does better with it The stomach is not rested by starvation The starvation treatment is now being generally abandoned except in the very severe cases as mentioned above With a turn of the wheel we are back where we were in 1904 when Lenhartz (24) introduced his diet for immediate feeding in hæmatemesis which was described in earlier editions of this book Meulengracht (13) has published figures showing a lower mortality rate than has been obtained by other methods of treatment and he advocates a liberal dietary from the first day of bleeding The routine he recommends is as follows 6 *a m* Tea white bread and butter 9 *a m* Oatmeal with milk white bread and butter 1 *p m* A selection from meat balls timbale broiled chops fish balls vegetable gratin fish gratin mashed potatoes vegetable purée vegetable soups cream of vegetables stewed apricots apple sauce gruel rice and tapioca puddings 3 *p m* Cocoa 6 *p m* White bread and butter sliced meats cheese and tea The patients are allowed as much as they like In addition one teaspoonful of the following powder is given Sod bicarb gr 30 mag carb lev gr 30 ext hyoscyam sicc gr 4 and ferri lact gr 5 t i d

We have tried this method with satisfactory results in a few cases but in others it has proved impracticable as the patient could not take the diet We doubt whether the true Meulengracht diet is now ever employed in this country the fact being that we can seldom stomach it There are indeed numerous modifications thereof which masquerade under the name of Meulengracht and cause much confusion Witts (22) recommends a diet which is more fluid than that of Meulengracht and contains no meat but which conforms to the principles of feeding by mouth on the first day of the hæmorrhage This has proved of great value in the treatment of many cases of hæmatemesis Thus Pickering (25) writing in 1942 says that during the last six years he has treated

one to two days Ice should not be sucked but attention must be given to the toilet of the mouth it being rinsed out from time to time with water (*d*) An hourly pulse chart should be kept (*e*) The blood pressure and hæmoglobin percentage should be determined every twelve to twenty four hours (*f*) The blood urea should be estimated and if raised determinations should be made daily until it falls to normal (*g*) The bowels should not be opened for four to seven days an enema then being given (*h*) Rectal salines of 6 to 8 oz with dextrose 2% should be given every four hours or a rectal drip may be used with a Murphy vulcanite rectal nozzle which allows free escape of flatus backwards along the tube to the reservoir as described by Marriott and Kekwick (21) Six to 8 pints of water normal or isotonic saline (0 85% sodium chloride) or isotonic dextrose (5% dextrose) may be run in slowly every twenty four hours resting the bowel every other hour by interrupting the flow (*i*) Blood transfusion is indicated according to Witts (22) if the hæmoglobin is below 40% the systolic blood pressure below 90 mm *Hg* the pulse rate over 140 or the blood urea over 100 mg per 100 ml Needham and McConachie (17) however found that in many patients who died the pulse rate did not rise as high as 120 The transfusion may be either a small one of 250 ml repeated as required or the drip method advocated by Marriott and Kekwick (23) may be used The amount of blood given by either method should raise the hæmoglobin to about 80% One pint of blood administered to an adult will increase the hæmoglobin by about 10% By the drip method this 10% increase should be achieved every four hours (*j*) The value of hæmostatics is open to question Five or 10 ml of Stypven containing 0 5 mg or 1 mg of Russell s viper venom may be given by mouth or 5 ml calcium gluconate (10%) intramuscularly once daily

After periods of one to two days and when the bleeding has been arrested as judged by absence of hæmatemesis and the general condition of the patient 4 oz of half strength normal saline (0 42% sodium chloride) should be given by mouth every four hours for twelve hours Milk feeds may now be begun first with milk 1 oz water 1 oz and emulsio mag oxid m 30 every two hours and then milk 2 oz every two hours and gradually increasing the milk feeds to 5 oz every two hours Tincture of belladonna (m 10–15) should be given before three feeds and olive oil (½ fl oz)

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found that not all the patients could take even his fluid diet from the start The diet is as shown on p 160

The milk is not citrated but may be flavoured with Horlick s malted milk or Ovaltine Vitamin C is given as fruit juice or as ascorbic acid tablet 50 mg t i d Sips of water or of dextrose solution or half strength normal saline are allowed between feeds up to 5 oz an hour The total fluid intake in the twenty four hours should be about 5 pints Neutralising substances or alkalis are only given if there is abdominal pain which is unusual No purgatives are given and if the bowels are not opened an enema is given on the seventh day Transfusion is given according to the criteria laid down above Woldman (26) treated 21 cases of hæmatemesis admitted to St Luke s Hospital Cleveland Ohio by the continuous intragastric aluminium hydroxide drip method and they all recovered In addition during the first twenty four hours the patients were given by mouth 2 oz of milk and cream every two hours and after that the diet included cooked cereals gelatine custard cream soup and rice and tapioca puddings Morphine was not used but barbiturates and other sedatives were administered as required Woldman considers the drip is a further advance on the principle of early feeding This compares favourably with the 38 cases of hæmatemesis admitted to the hospital during the previous five years who were not treated by the drip for 11 of these cases died

The Surgical Treatment of Hæmatemesis Gordon Taylor (27) urges that the surgeon should not be excluded from consultation until the last desperate stage of ulcer hæmorrhage and supports the view expressed by Finsterer (28) that the first forty eight hours is the optimum period for operation The practical difficulty is to decide in any individual case whether death will occur if medical treatment is adopted and whether life could be saved by an early operation Further it is often impossible to decide at this stage whether the patient has an acute or chronic ulcer or chronic gastritis The chief indications for operation are (*a*) The erosion of a large vessel (*b*) Concomitant hæmorrhage and perforation of the stomach (*c*) Hæmorrhage which cannot be controlled by blood transfusion (*d*) A primary hæmorrhage may require surgical treatment in elderly people whose vessels are thickened and blood pressure raised Needham and McConachie

all his patients (over 40 in number) suffering from gastric and duodenal hæmorrhage by the diet recommended by Witts with only one death and he supports Witts' view that "the results of

DIETETIC TREATMENT OF GASTRO-DUODENAL HÆMORRHAGE ARRANGED FOR TWO-HOURLY FEEDING

Feeds by Day	Food		Day 1	Day 2	Day 3 and subsequent
1	Whole milk (fresh or dried)	oz	5	5	5
	Patent barley or strained porridge		Portion	Portion	Portion
2	1 egg beaten up in milk	oz	5	5	5
	Buttered rusks or cream crackers		–	1	2
3	Whole milk (fresh or dried)	oz	5	5	5
	Marmite to taste		–	–	–
	Barley sugar	oz	1	1	1
	Thin crustless white bread and butter slices		–	1	2
4	Strained orange or tomato juice	oz	1	1	1
	Vegetable purée		Portion	Portion	Portion
	Pudding				
	Cream	oz	1	1	1
	Boiled or steamed fish		–	–	Portion
5	1 egg beaten up in milk	oz	5	5	5
	Barley sugar	oz	1	1	1
	Buttered rusks or cream crackers		–	1	2
6	Whole milk (fresh or dried)	oz	5	5	5
	Fruit purée		Portion	Portion	Portion
	Pudding				
	Cream	oz	–	1	1
	Thin crustless white bread and butter slices		–	1	2
7	1 egg beaten up in milk	oz	5	5	5
	Black treacle or barley sugar	oz	1	1	1
	Buttered rusks or cream crackers		–	1	2
8	Whole milk (fresh or dried)	oz	5	5	5
	Fruit purée		Portion	Portion	Portion
	Pudding				
Feeds at night (when awake)	1 Whole milk (fresh or dried)	oz	5	5	5
	2 1 egg beaten up in milk	oz	5	5	5
Between feeds	Strained orange or tomato juice	oz	1	1	1
	Approximate calorie value		2 545	3 118	3 0[illegible]4

immediate feeding supplemented when necessary by transfusion are much superior to starvation or operation." The advocates of early feeding to the exclusion of the mouth starvation method must, however, remember that in Witts' original series of cases he

owing to its expense to the slaty colour it gives to the fæces which interferes with the detection of blood and because it is said neither to relieve pain nor to neutralise acidity Clinically however bismuth undoubtedly exerts a sedative effect Magnesium oxide and magnesium hydroxide are both powerful neutralising agents Sodium and potassium citrate tribasic calcium phosphate and tribasic magnesium phosphate neutralise hydrochloric acid without producing an alkaline solution (30 31) Crohn (32) in 1929 introduced colloidal aluminium hydroxide as an antacid Six daily doses of 60 to 120 m of a preparation such as Aludrox (5 8 / aluminium hydroxide) in water may be given between feeds Fauley et al (33) have shown that aluminium hydroxide gel in relatively large doses interferes with the absorption of phosphates in man and dog and that it may produce a phosphorus deficiency in the presence of a relative deficiency of pancreatic juice of diarrhœa or of a low phosphorus diet This is however not likely to occur with a patient on an ordinary ulcer diet They also showed that aluminium phosphate gel will prevent the formation of a peptic ulcer in Mann Williamson dogs (in which a gastro jejunostomy is performed and the pancreatic juice and bile are diverted into the last 20 to 25 cm of the ileum) In such dogs a jejunal ulcer develops with death in about eleven weeks Aluminium phosphate gel does not interfere with phosphate absorption Aluminium hydroxide gel did not prevent ulcer formation in these dogs The use of aluminium phosphate gel in a few cases of patients suffering from peptic ulcer indicated that it was as effective as any other neutralising agent and it is probably preferable to aluminium hydroxide gel in cases asssociated with pancreatic deficiency or diarrhœa Mutch (34) has obtained good results with a synthetic hydrated magnesium trisilicate in doses of gr 5 to 21 (anhydrous weight) between each feed It possesses antacid and general adsorbent properties Cohn et al (35) record the case of a granuloma of the stomach following the ingestion of a medication containing hydrated aluminium silicate which the patient had been taking for a long time for a duodenal ulcer

There is some risk of producing alkalosis if large doses of magnesia or sodium bicarbonate are given but the risk is diminished if magnesium trisilicate or colloidal aluminium hydroxide are employed

(17) consider that surgical treatment is indicated in the majority of patients over 50 who suffer a further hæmorrhage after admission to hospital An operation is rarely if ever advisable in primary hæmorrhage under other circumstances and acute ulceration of the stomach probably never requires surgical treatment

The position with recurrent hæmorrhages is somewhat different Previous X ray investigation will probably have been carried out With a large penetrating ulcer a fibrotic ulcer with pyloric or duodenal stenosis or with mid gastric narrowing an operation may be required to prevent further bleeding It should not be performed until the hæmoglobin has been raised to 60% by blood transfusion In no case should an operation be advised unless very skilled surgery is available with adequate facilities for blood transfusion for the mortality rate for operation for active hæmorrhage from chronic ulcer is at the best about 10% In all cases in which the patient is under forty five years of age an operation is best avoided

The Medical Treatment of Ulcers not Complicated by Hæmorrhage The modified Sippy treatment is now usually employed

The Sippy Treatment (modified) or The Alkali Treatment Sippy (29) of Chicago introduced this treatment in 1915 Its object is to protect the ulcer from hydrochloric acid This is accomplished by giving large doses of alkalis after and of belladonna before the feeds and by using as the basis of the diet milk and cream

Modifications of the Sippy treatment have been found to yield good results and are more easily carried out than the original method The feeds are given at two or two and a half hourly intervals an alkaline powder or neutralising substance is taken one hour after and olive oil or belladonna before the feeds There is some difference of opinion as to the best alkalis or neutralising substances for the purpose Sippy (29) recommended sodium bicarbonate heavy magnesia and bismuth carbonate All carbonates have the disadvantage of liberating CO_2 and causing gaseous distension which proves distressing in some cases Sodium bicarbonate and magnesium oxide provoke a secondary secretion of gastric juice Bismuth carbonate has no neutralising power and theoretically is of little value as it is improbable that it forms a protective coat to the ulcer It is however out of fashion now

owing to its expense to the slaty colour it gives to the fæces which interferes with the detection of blood and because it is said neither to relieve pain nor to neutralise acidity Clinically however bismuth undoubtedly exerts a sedative effect Magnesium oxide and magnesium hydroxide are both powerful neutralising agents Sodium and potassium citrate tribasic calcium phosphate and tribasic magnesium phosphate neutralise hydrochloric acid without producing an alkaline solution (30 31) Crohn (32) in 1929 introduced colloidal aluminium hydroxide as an antacid Six daily doses of 60 to 120 m of a preparation such as Aludrox (5 8 % aluminium hydroxide) in water may be given between feeds Fauley et al (33) have shown that aluminium hydroxide gel in relatively large doses interferes with the absorption of phosphates in man and dog and that it may produce a phosphorus deficiency *in the presence of a relative deficiency of pancreatic juice of* diarrhœa or of a low phosphorus diet This is however not likely to occur with a patient on an ordinary ulcer diet They also showed that aluminium phosphate gel will prevent the formation of a peptic ulcer in Mann Williamson dogs (in which a gastro jejunostomy is performed and the pancreatic juice and bile are diverted into the last 20 to 25 cm of the ileum) In such dogs a jejunal ulcer develops with death in about eleven weeks Aluminium phosphate gel does not interfere with phosphate absorption Aluminium hydroxide gel did not prevent ulcer formation in these dogs The use of aluminium phosphate gel in a few cases of patients suffering from peptic ulcer indicated that it was as effective as any other neutralising agent and it is probably preferable to aluminium hydroxide gel in cases asssociated with pancreatic deficiency or diarrhœa Mutch (34) has obtained good results with a synthetic hydrated magnesium trisilicate in doses of gr 5 to 21 (anhydrous weight) between each feed It possesses antacid and general adsorbent properties Cohn et al (35) record the case of a granuloma of the stomach following the ingestion of a medication containing hydrated aluminium silicate which the patient had been taking for a long time for a duodenal ulcer

There is some risk of producing alkalosis if large doses of magnesia or sodium bicarbonate are given but this risk is diminished if magnesium trisilicate or colloidal aluminium hydroxide are employed

Milk is the best neutralising food substance and the addition of sodium citrate gr 3 to the ounce not only prevents curds forming but also increases the neutralising effect Fatty substances tend to diminish gastric secretion carbohydrates have little neutralising power

During the stage of healing of the ulcer the patient must be kept in bed and feeds given at not more than two to two and a half hourly intervals during the day by night a feed should only be given if the patient wakes The time for an ulcer to heal as judged by X ray appearances and negative tests for occult blood in the fæces varies according to its size from four to eight weeks It is improbable that a deeply excavated and adherent ulcer could heal so quickly Although some authorities maintain that it is irrational to increase the diet until the ulcer is healed in our experience this can be done with advantage by carefully graduated changes

The treatment we adopt is as follows —

Healing Stage (Diet 1) *Weeks 1 and 2* Feeds of 5 oz are given at 7 a m 9 a m 11 a m 1 p m 3 p m 5 p m 7 p m 9 p m 11 p m and once during the night if awake The feeds consist of milk (warm and containing 3 gr soldium citrate to the ounce) Horlick s malted milk or Benger s food At least five of the feeds should be of milk which may be coloured with tea Sugar may be added to the feeds as desired If the patient is hungry the feeds may be increased to 6 oz or even 8 oz and ½ oz of glucose taken 3 times a day Strained orange or tomato juice 1 oz should be given daily thoughout the treatment to supply vitamin C

Week 3 Add one raw egg to two of the milk feeds if available A little thin bread and butter is given with one feed and a rusk or plain biscuit may be taken with the other feeds In addition the patient may be given once a day a little of one of the following sweet jelly milk jelly custard or junket The feeds are still given two hourly

Week 4 The feeds are given every two and a half hours from 7 a m to 10 p m They consist of 5 oz of citrated milk every other feed with one egg in two of the feeds if available the thin bread and butter rusk or plain buscuit and sugar as in week 3 The alternate feeds are composed of a similar quantity of potato soup arrowroot cornflour or milk pudding (sago or tapioca) and once

a day a little sweet jelly milk jelly custard junket apple or prune puree or honey

Week 5 The feeds are given every two and a half hours The milk may now be reduced to three times a day at 7 a m 5 p m and 10 p m Additions to the diet are a small helping of porridge made from very fine oatmeal pounded fish 2 oz lightly boiled egg crisp toast or rusk (well chewed) with butter sponge or Madeira cake

Week 6 Feeds still every two and a half hours Add 2 to 4 oz minced meat daily

Take immediately before three feeds $\frac{1}{2}$ oz of olive oil and immediately before three other feeds tinct bellad m 5–10 aq chlorof ad fl oz $\frac{1}{2}$ or atrophine sulphate gr $\frac{1}{100}$ in m 60 of water Take one teaspoonful of the magnesium trisilicate or of Aludrox one hour after five feeds during the day and two teaspoonfuls the last thing at night in a little water during the first two weeks of the treatment

During the third and fourth weeks take a teaspoonful of the powder or of Aludrox three times a day after feeds and two tea spoonfuls at night In the fifth to eighth weeks take a teaspoonful of the powder or of Aludrox twice a day after feeds and two teaspoonfuls at night It is very important to neutralise as far as possible the gastric secretion during the night The patient should always have a milk feed by his bed to drink if he wakes The mouth should be cleaned after each feed with bicarbonate of soda solution gr 60 to 5 oz of water This is important as parotid suppuration may occur if it is neglected No smoking is allowed

Convalescent Stage (Diet 2) During this period of about a month the patient is allowed up for gradually increasing periods and additions are made to the dietary

7 30 *a m* *Breakfast* One egg (lightly boiled if available) or 2 oz of steamed fish thin bread and butter or crisp toast buttered when cold (to be well chewed) honey or apple jelly sugar and milky tea

10 *a m* Milk 5 oz (containing 15 gr of sodium citrate) or Horlick s malted milk or Benger s food

12 30 *p m* *Lunch* Milk soup (potato or artichoke) fresh white fish (boiled or steamed) or rabbit chicken or tender mutton mashed potato Custard or milk pudding or stewed apples (with

no pips skin or core) or prune purce and the juice of an orange if available

3 p m Thin bread and butter or biscuits and butter or one small slice of sponge or Madeira cake and milky tea

5 30 p m Milk 5 oz (containing 15 gr sodium citrate) or Horlick's malted milk

8 p m *Dinner* As for lunch

10 30 p m Milk 5 oz (containing 15 gr sodium citrate) or Benger s food

The neutralising powder or Aludrox should be taken one tea spoonful after breakfast and two teaspoonfuls last thing at night and the olive oil or belladonna before breakfast lunch and dinner The meals should be small and eaten slowly No smoking is allowed

After Treatment (Diet 3) *8 a m* *Breakfast* A selection from porridge made from very fine oatmeal egg boiled poached or scrambled white fish grilled or steamed cold ham tongue toast crisply made and well chewed breakfast biscuits honey apple jelly butter sugar and milky tea or coffee

11 a m A tumbler of milk Ovaltine Horlick s malted milk or Benger s food and a biscuit

1 p m *Lunch* A selection from soup made from milk and vegetables such as potato or artichoke Fresh white fish boiled grilled or steamed with white sauce or fish soufflé Meat such as minced fresh beef mutton rabbit liver or veal grilled lamb cutlet, boiled or roast chicken If the meat is not minced it must be well masticated Cheese soufflé Vegetables such as mashed potato steamed or boiled green vegetables marrow peas (if tender) Sweets such as custard boiled or baked junket milk pudding very well cooked cornflour stewed apples (with no pips core or skin) chocolate soufflé plain steamed or baked pudding Crisp toast butter and the juice of one or more oranges

4 p m A cup of milky tea thin bread and butter jelly jam or honey, sponge or plain cake

7 p m *Dinner* As for lunch

10 p m A feed as at 11 a m

The breakfast lunch and dinner should be small meals well chewed and eaten slowly The patient should rest for half an hour after meals No condiments should be eaten and nothing taken which cannot be reduced to a soft pulp in the mouth If it is

impossible to obtain the liquid feeds between all the meals a biscuit or some chocolate should be eaten A teaspoonful of the neutralising powder or of Aludrox should be taken every night on retiring to bed No smoking is allowed

The following articles of food are forbidden Smoked salmon kippers whitebait tough meat pork high game sausages brawn curry made up dishes fried foods cheese (except cream cheese) meat extracts and meat soups pickles spices ginger salads uncooked vegetables celery carrots cucumber onions or any fibrous vegetables new or wholemeal bread buns Ryvita Vita wheat digestive and oatmeal biscuits hot buttered toast unripe or raw fruit rhubarb plums nuts raisins sultanas figs jam with pips marmalade with peel strong tea or coffee aerated drinks and alcohol

The Danger of Alkalosis Large doses of alkalis especially of sodium bicarbonate magnesium oxide and carbonate may *produce toxic symptoms (Hardt and Rivers (36))* *Alkalosis* is liable to occur in cases of gastric stasis especially associated with duodenal ulcer or renal insufficiency It would appear that this observation is difficult to reconcile with the rationale of the alkali treatment of nephritis for renal insufficiency usually tends to produce acidosis The symptoms are headache vertigo loss of appetite distaste for milk malaise nausea vomiting drowsiness or coma A determination of the alkali reserve of the blood is of value in confirming the diagnosis All alkalis should be stopped dextrose given by mouth or in rectal salines ammon chlor 0 5 g (stearettes) should be administered by mouth every six hours or by rectum in doses of 4 g every six hours and an intravenous injection made of collosol calcium (1 ml) repeated next day if necessary

Continuous Intragastric Drip Treatment Winkelstein (37) in 1932 determined the gastric acidity during the night in cases of duodenal ulcer gastric ulcer and normal controls A Rehfuss tube was inserted in the stomach at 7 p m and specimens of gastric juice removed every two hours during the night He found a high continuous acid secretion in cases of duodenal ulcer a moderate acid secretion in gastric ulcer but in the controls the curve was very low and in 4 out of every 6 cases no free hydrochloric acid was secreted during the night He introduced a continuous Murphy drip through a Rehfuss tube for the treatment of peptic

no pips skin or core) or prune purée and the juice of an orange if available

3 p m Thin bread and butter or biscuit and butter or one small slice of sponge or Madeira cake and milky tea

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8 p m Dinner As for lunch

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The breakfast lunch and dinner should be small meals well chewed and eaten slowly The patient should rest for half an hour after meals *No condiments* should be eaten and nothing taken which cannot be reduced to a soft pulp in the mouth *If it is*

The Medical Treatment of Chronic Ulcers This is in all essentials similar to that detailed above for acute ulceration of the stomach or duodenum in fact apart from the history of the duration of the case it is difficult to distinguish between the two conditions

It will be noticed that throughout this section gastric and duodenal ulcers have been considered together as if their treatment were the same Duodenal ulcers are of much more frequent occurrence and are more easy to diagnose

The Surgical Treatment of Ulcers not Complicated by Hæmorrhage While on the one hand it is clear that in certain circumstances medical measures are of no avail in the treatment of gastric and duodenal ulcers yet patients are frequently seen who have had operations for these conditions and who have received no permanent benefit Indeed their last state is not infrequently worse than their first It is therefore profitable to endeavour to decide what are the indications for surgical treatment In this way patients should derive the maximum benefit possible and not be subjected to operations which either give no relief or are followed by only a temporary amelioration of the symptoms

The following are the chief indications for operations —

Surgical treatment is imperative in cases of perforation or of perigastric abscess although some cases of perforation have been successfully treated by continuous gastric suction

Ogilvie (41) makes the following further recommendations —

Gastric Ulcer Operation is required for the following reasons (*a*) Organic stenosis causing delay in emptying (*b*) If cancerous transformation of the ulcer is suspected (*c*) If medical treatment fails as will occur if a chronic ulcer becomes fixed to neighbouring structures The acid secretion in gastric ulcer is not usually raised and gastrectomy is generally the most satisfactory operation

Duodenal Ulcer Operation is required for organic stenosis A posterior gastro enterostomy or a gastro duodenostomy should be performed As the gastric acidity is usually low owing to the presence of chronic gastritis recurrent anastomotic or jejunal ulceration is unlikely to ensue Recurrent ulceration of this type is probably due to the operation of gastro enterostomy being performed in the presence of high gastric acidity Further the average time for the appearance of the recurrent ulcer is several years Gastro jejunostomy alone is therefore absolutely contra

ulcer using alkalinised milk Woldman and Rowland (38) in 1936 published their results in cases of peptic ulcer treated with a continuous intragastric drip of 200 ml of 7% colloidal aluminium hydroxide mixed with 600 ml of water These authors (39) reported further that gastric and duodenal ulcers which were quite apparent radiologically disappeared after about seven to fourteen days treatment further only 3 or 4 small bland feeds a day were required such as milk cream junket custard jelly cereals etc Rutherford and Emery (40) in 1939 and 1940 used Creamalin (5 5% aluminium hydroxide) 1 part to 3 parts of water as a drip running at 15 drops a minute They treated 28 severe cases of peptic ulcer which had previously failed to respond to medical or surgical treatment or to both They gave the drip usually for a week and subsequently the aluminium hydroxide was administered by mouth for several months In a follow up fifteen months later of 14 of their original 28 very severe cases they reported that 8 were well 2 doubtfully improved and 4 were unsatisfactory We use a mixture of milk 1 pint dextrose 2 oz and sod citrate gr 10 A Ryle s tube is passed through the nose into the stomach and joined by a glass connection to rubber tubing attached to the drip *The fluid flows at the rate of 40 drops a minute* day and night *i e* 6 pints in twenty four hours If the tube blocks it is flushed through with a syringeful of milk A specimen is removed every four hours for the first twenty four hours and titrated for free HCl if it is present ½ fl oz of Aludrox is added to each pint of milk If the patient is hungry the amount of dextrose is increased The Ryle s tube is changed every other day cleaned and boiled *and the rubber tubing and glass connections are* washed and boiled We have found that some cases of gastric or duodenal ulcer which have not healed after six to eight weeks treatment in bed on the conventional Sippy lines have healed after treatment by the drip method in periods of three to seven weeks

Whatever form of treatment is adopted it is important that as soon as the patient is in a fit state a search should be made for any *possible focus of infection* which should be eradicated when found The teeth should be X rayed the nose and sinuses examined and pyorrhœa should be treated A search for signs of appendicitis or infection of the gall bladder or uro genital tract should also be made

per twenty four hours to 300 to 600 ml per twenty four hours and gastric ulcers did not develop The gastric acidity was also lowered It was this experiment which led Dragstedt to divide the vagi in man for the treatment of peptic ulcer Since then vagotomy has been more extensively used as a therapeutic measure especially as so much emphasis is now laid on the effect of nervous stress emotion and frustration in the production of peptic ulcers Vagotomy results in the abolition both of gastric hypersecretion and hypermotility and gastric acidity and secretion tend to return to normal

In 1946 Dragstedt (46) reported very promising results from vagotomy in the treatment of peptic ulcer and observed no recurrence of ulcers although he cautiously stated a longer period of observation and a more extensive series of patients are obviously necessary to provide final answers Thorek (47) reviewed in 1947 the anatomical nerve supply of the stomach and emphasised the relative merits of the transabdominal and transthoracic methods of approach He concluded that the operation appeared to be indicated in duodenal and stomal ulcers which do not respond to medical treatment He advised that gastric ulcers should be resected owing to their tendency to malignant degeneration The transabdominal operation has the advantage that the lesion can be inspected and a gastro enterostomy performed to compensate for obstructive or atonic complications It has been shown by many observers that if all the vagal fibres are not divided the gastric acidity tends to return to its previous values Thompson and James (48) believe the vagi can be more completely divided by the transthoracic approach They report satisfactory results in 4 patients with severe and recurrent peptic ulceration Orr and Johnson (49) have tried vagotomy as an alternative to gastro jejunostomy which they state is a failure as a routine procedure for it does not cure more than half the patients and has an operative mortality rate of about 2% Gastrectomy they say is more successful if sufficiently radical but has a mortality rate of about 5% and a recurrent ulceration rate of 2 to 9% These surgeons performed vagotomy or vagal resection on 50 patients without a death The operation they prefer is post hiatal vagal resection from below A gastro enterostomy may subsequently be necessary if the stomach does not recover sufficient tone to drive the meal through the narrowed

indicated when the gastric acidity is high and the rate of stomach emptying is rapid with the following exception In a patient over the age of fifty if symptoms of duodenal ulcer persist usually because the patient will not adhere strictly to his medical treatment a short circuiting operation may be performed even if there is no stenosis The gastric acidity tends to fall progressively after the age of fifty and so the chance of a recurrent ulcer is slight

Vagotomy

Division of the vagus nerves to eliminate gastric secretion due to nervous stimuli would appear to be a sound procedure in cases of peptic ulcer as such ulceration is believed to result from the corroding effect of unneutralised gastric hydrochloric acid Vagotomy will not influence the gastric secretion which results from hormonal influences and which accompanies the taking of food The presence of the meal in the stomach however probably protects the gastric mucosa from the effects of the hydrochloric acid The nervous secretion occurs as the result of psychic stimulation at the thought or sight of a meal and also accounts for the considerable secretion of gastric juice which is poured out during the night By diminishing or abolishing such nocturnal secretion vagotomy should exert a powerful influence both in the healing of peptic ulcers and in preventing their recurrence

As far back as 1814 Brodie (42) showed that the intravenous injection of arsenic in dogs causes the stomach to be filled with mucus and watery fluid If the vagi were cut before the arsenic was injected the stomach remained empty In 1894 Pavlov (43) found that division of the vagi in dogs prevented the gastric secretion of hydrochloric acid In 1923 Carlson (44) pointed out that although vagotomy abolished the neurogenic secretion there still remained the humoral secretion due to the formation of gastrin in the pyloric mucosa which is carried by the blood stream to the stomach In 1930 Dragstedt and Ellis (45) isolated the stomach in dogs keeping the blood and nerve supplies intact and maintaining intestinal continuity by an oesophago jejunostomy The gastric secretions were collected through a gold plated cannula Chlorides had to be supplied to keep the animals alive In such animals gastric ulcers developed When the vagi were divided the amount of gastric secretion was reduced from 1 000 to 2 ,00 ml

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recurrent ulcer symptoms Beattie had to operate again on 34 of his 103 cases of simple vagotomy to relieve retention symptoms In order to prevent these Beattie advises performing pylorectomy together with vagotomy and considers that gastrectomy should be reserved for those less common cases in which the ulcer has penetrated deeply into the surrounding viscera

Conclusions Vagotomy offers a physiological method of reducing the gastric acidity which leads to the formation of peptic ulcers and is now being more extensively used especially when combined with some other operation The disturbances of motor function and the tendency to the secondary development of pyloric spasm may lead to a recurrence of ulcer symptoms causing considerable distress and requiring a subsequent gastro enterostomy It is not yet possible to say whether vagotomy will become the operation of choice in the majority of cases of duodenal ulcer but it does seem probable that vagotomy should be combined with some other procedure to eliminate the probable delay in stomach emptying Although the physician should understand what types of operation are available for the treatment of peptic ulcer the ultimate choice is the responsibility of the surgeon

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pyloru They consider that vagal resection is particularly indicated in young subjects who suffer from relapses of duodenal ulcer whereas gastrectomy is the operation of choice for gastric ulcer Vagal resection may also be used with advantage in cases of stomal ulcer, either with or without other operative procedures

The insulin test is of value in determining whether the vagal fibres have been adequately divided With an intact vagal supply the intravenous injection of 16 units of insulin causes a sharp rise in the free and total acidity of the gastric juice This is due to hypoglycæmia affecting the vagal nerve centre and is not a primary insulin effect After vagal resection no such rise occurs

Walters et al (50) in 1948 reported the results of vagus nerve resection in 118 cases of gastro duodenal and jejunal ulceration The authors operated on patients suffering from duodenal ulcer, when treatment had failed Such cases they would formerly have considered suitable for partial gastrectomy Vagotomy was combined with gastro enterostomy when indicated Relief of pain and healing of the ulcer is much more likely to occur with duodenal than with gastric ulcer Rob (51) has recounted a case of recurrence of symptoms of duodenal ulcer six months after complete freedom of symptoms following bilateral vagotomy The insulin test indicated that the vagal resection was adequate and there was no evidence of nerve regeneration when the symptoms recurred Rob found that in a series of 37 operations for peptic ulcer the results were very good in 5 cases with secondary ulceration following gastrectomy or gastro enterostomy In the other 32 cases with duodenal ulcer there were numerous early complications such as temporary severe dysphagia persistent diarrhœa and eructations of foul gas Beattie (52) reviewed in 1950 the results of gastrectomy and vagotomy in duodenal ulcers The after results of gastrectomy for duodenal ulcers are often disappointing He says that the stomach is too small to hold a sufficiency of food or to digest it properly and recurrent gastro intestinal upsets occur in about a sixth of the cases which are sufficiently severe to upset the patient s economic life In a series of 3 000 successful vagotomies performed during an eight year period up to 1950 it was found that recurrent ulceration is extremely rare when the insulin test has shown complete nerve section Secondary pylorospasm due to over action of the sympathetic nerves however frequently caused

CHAPTER VIII

THE CARDIOVASCULAR SYSTEM

Cardiac Irregularities

Potain (1) in 1867 obtained tracings from the jugular vein in man He used as a receiver of its pulsations a glass funnel which was connected by a rubber tube with a Marey sphygmograph and recorded at the same time tracings from the apex beat carotid and radial arteries In 1892 Sir James Mackenzie (2) using a small metal cup as a receiver devised the instrument originally called a phlebograph by means of which simultaneous tracings of the pulsations of the jugular vein and radial artery were obtained The records were taken upon smoked paper but by the use of inked pens the polygraph was evolved

Our knowledge of the essential nature of the different forms of cardiac irregularity is mainly due to Sir James Mackenzie s pioneer work with this instrument by means of which he obtained records representing the activities both of the auricle and of the ventricle Further light has been thrown upon this subject and upon myo cardial disorders by the use of the electrocardiograph

The chief varieties of cardiac irregularity are as follows (1) Sinus arrhythmia (2) premature systoles (3) heart block (4) auricular flutter (5) auricular fibrillation (6) paroxysmal tachycardia and (7) pulsus alternans

The Electrocardiograph

The electrical currents produced by the contraction of the human heart were first recorded by Waller (3) in 1887 by means of the capillary electrometer With this instrument the movements of a fine column of mercury are photographed on a moving plate

The string galvanometer was later introduced by Einthoven (4) in 1903 and modifications of it are now known as the electro cardiograph

Immersion electrodes have been largely replaced by small electrodes of German silver plate about 3 square inches in size Cambridge electrode jelly is smeared over the electrode and rubbed

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(51) ROB *Lancet* 1948 ii 730
(52) BEATTIE *Lancet* 1950 i 525

Certain time relations are important —

The P–R interval is normally 0 14 second Prolongation of this indicates delay in conduction in the A–V node or the upper part of the bundle of His

The QRS interval is normally just under 0 10 second Prolongation of this occurs in the various grades of heart block and in ectopic ventricular beats

The R–T interval is normally 0 32 second Prolongation of this occurs especially in complete A–V block

The QRST curves are produced by currents originating in the two ventricles and represent the algebraic sum of the waves produced by the two ventricles contracting separately

The characteristic appearance of the deflections in the first three leads should now be studied (see Fig 9) The R wave is normally greatest in Lead II and R_1 and R_3 are together approximately equal to R_2 (The figures below the waves indicate the leads in which the waves occur) The P and T waves are also usually larger in Lead II than in Leads I or III

The Significance of certain Alterations in the Form of the Deflections *The P Wave* This may be inverted indicating that the impulse originates at a new focus in the auricle (at times in the A–V node as in paroxysmal tachycardia and in nodal extrasystoles) The P wave may be absent or replaced by a small series of oscillations of a fine or coarse variety as is seen in auricular fibrillation It may be abnormally large and notched indicating auricular hypertrophy as may occur in mitral stenosis P_3 may be greater than P_2 as may be seen in auricular flutter

The R Wave A maximum R wave in Lead I indicates left axis deviation which when present to a high degree is most often due to left ventricular preponderance It may also be associated with an unduly transverse lie of the heart associated with a high diaphragm In right axis deviation which also includes right ventricular preponderance the R wave is greatest in Lead III

Normally the left ventricle is 1 8 times heavier than the right

The S Wave A maximum S wave in Lead I indicates right axis deviation whereas a maximum S wave in Lead III is a sign of left axis deviation

The T Wave It is held that the T wave is of great importance as indicating the condition of the ventricular muscle A negative

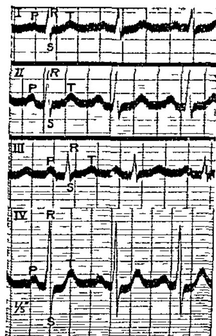


Fig 9 Normal Electrocardiogram (Lead IV R left arm electrode anteriorly right arm electrode on right arm)

gently into an area of skin corresponding with the size of the electrode

The leads usually adopted are as follows —

Lead I (transverse) Right and left hands

Lead II (axial) *Right hand and left foot*

Lead III (left lateral) Left hand and left foot

Lead IV (chest leads) See p 189

Unipolar leads See p 190

Curves representing the changes in potential caused by the heart s beat are photographed A single beat from the normal heart shows the following waves (see Fig 9) —

P is due to auricular systole and constitutes *the auricular* complex **QRST** occur during ventricular systole and constitute the ventricular complex **Q** and **S** are often absent **T–P** represents diastole

Certain time relations are important —

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Normally the left ventricle is 1 8 times heavier than the right

The S Wave A maximum S wave in Lead I indicates right axis deviation whereas a maximum S wave in Lead III is a sign of left axis deviation

The T Wave It is held that the T_2 wave is of great importance as indicating the condition of the ventricular muscle A negative

T_2 wave is always pathological and a negative T_1 wave usually indicates a pathological condition. With full doses of digitalis the S–T segment is usually depressed and the T wave inverted in all leads. A negative T_3 wave is not necessarily pathological.

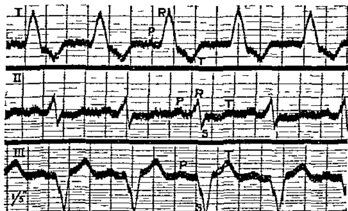


FIG. 10. Bundle Branch Block (Type I)

Prolongation of the QRS Complex. In the condition known as bundle branch block, the right or left branch of the bundle of His is impaired and fails to conduct. The QRS complex is prolonged or spread out, and lasts longer than the normal maximum of 0.10 second. The R wave may be notched. The T wave is in the opposite direction to the main initial deflection (R or S) and is usually of a greater amplitude than normal. The ventricular complex is therefore diphasic.

Two types of curves are met with in patients suffering from bundle branch block, depending on which branch of the bundle is obstructed. The old classification was based on animal experiments and according to the type of curve obtained right or left bundle branch block was diagnosed. Later work on human subjects has thrown doubt on the applicability to man of these conclusions drawn from animal experiments and there is therefore uncertainty as to which type of curve corresponds with right or left bundle branch block. The curves are referred to now simply as Type I and Type II.

Type I Bundle Branch Block (old terminology right). The main

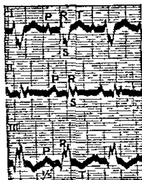


FIG 11 Bundle Branch Block (Type II)

initial deflection (R) is positive in Lead I and negative (S) in Lead III (*see* Fig 10) This is the common variety

Type II Bundle Branch Block (old terminology left) The main initial deflection (S) is negative in Lead I and positive (R) in Lead III (see Fig 11) **Type IIa** This was described by Wilson and his co workers (5) in 1934 and in 1937 was called by Evans and Turnbull (6) the newer electrocardiogram denoting right bundle branch block It is more common than the hitherto standard Type II curve The distinctive features are Lead I R and S almost equal in size R narrow S broad and notched T upright Lead II S broad and deep T usually upright Lead III Q or S usually deep T inverted (see Fig 12)

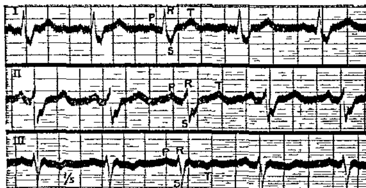


FIG 12 Bundle Branch Block (Type IIa)

The QRS complex is also prolonged in *intraventricular block* in which the terminations of the bundles in the ventricular sub endothelial tissue are destroyed The R wave is also often notched and the T wave is in the same direction as the initial main deflection

In *congenital dextrocardia* all the waves in Lead I are inverted whereas Lead II may represent the normal Lead III, and vice versa

Absence of the PRT Complex This occurs in sino auricular block, in which the stimulus arising in the S–A node at times fails to cause a contraction of the auricle The whole heart therefore misses a beat but the succeeding contraction occurs at approximately the normal interval This is not necessarily a serious condition (see Fig 13)

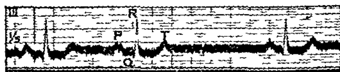


Fig 13 Sino-Auricular Block

In addition to the light which the electrocardiograph throws upon the condition of the myocardium and conducting tissue the various types of cardiac irregularity can be accurately determined

Sinus Arrhythmia Variations in the length of the T–P intervals are seen but the auricular and ventricular complexes are normal

Premature Systoles *Auricular* The premature impulse may arise at the S–A node near it or some distance away In the two former instances the P wave caused by the premature contraction is in the normal direction in the latter it is inverted indicating that the impulse spreads through the auricle along abnormal

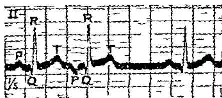


Fig 14 Premature Auricular Systole

paths (see Fig 14) The ventricular complex following the premature auricular stimulus is usually of normal type The premature P wave may occur so early in diastole that it fuses with the previous T wave

Usuall) the diastole followmg the premature beat is not fully compensator)



FIG 1 Premature Nodal Systole

Nodal The premature impulse arises in the junctional tissue uniting the auricle and ventricle *i e* in the A–V node or the A–V bundle before division The ventricular contraction may occur just before or abnormally shortly after the auricular but is seen to be supra ventricular in origin because the QRS complex is of normal appearance (see Fig 15)

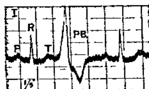


FIG 16 Premature Ventr cular Systole

Ventricular A premature stimulus arises in the ven tricle below the division of the main A–V bundle The premature ventricular com plex resulting has the same duration as a normal ven tricular complex but its form is altered the initial deflec tions of the premature beat being larger than normal

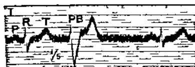


FIG 17 Premature Ventricular Systole

The auricular waves occur at regular intervals and may be fused with the premature venticular deflections or appear just after them The premature ventricular beat is followed by a prolonged diastole and the succeeding pause is thus fully compensatory

Two types occur (see Figs 16 and 17) formerly called left and right ventricular premature systoles As with the site of bundle branch block the localisation of ventricular systoles is now a matter of doubt but of little clinical significance According to the old terminology based on experiments on dogs the main ventricular deflection is upright in Lead I and inverted in Lead III with left

In *congenital dextrocardia* all the waves in Lead I are inverted whereas Lead II may represent the normal Lead III and vice versa

Absence of the PRT Complex This occurs in sino auricular block in which the stimulus arising in the S-A node at times fails to cause a contraction of the auricle The whole heart therefore misses a beat but the succeeding contraction occurs at approximately the normal interval This is not necessarily a serious condition (see Fig 13)



Fig 13 Sino Auricular Block

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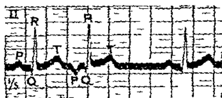


Fig 14 Premature Auricular Systole

paths (see Fig 14) The ventricular complex following the premature auricular stimulus is usually of normal type The premature P wave may occur so early in diastole that it fuses with the previous T wave

occur (see Fig 13) If the rate is slow as is usually the case on exercise it may increase to about double

When the heart is beating at such a slow rate the ventricle may interpolate a beat on its own (*ventricular escape*) the P–R interval being shortened or the P and R waves may partially fuse

Nodal Rhythm The impulse may arise at the A–V node instead of at the S–A node the auricles and ventricles contract simultaneously and no **P** waves are seen or they occur shortly after the R–S complex and are usually inverted (see Fig 19)

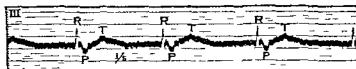


Fig 19 Nodal Rhythm.

Auricular Flutter An electrocardiogram shows the auricular contractions occurring at a regular rate with ventricular complexes superimposed upon the curve at regular or irregular intervals Heart block of varying grades is almost invariably present

The **P** waves occur at regular intervals and have a dome shaped appearance (see Fig 20) In some tracings the **T** wave is also apparent



Fig 20 Auricular Flutter

Pressure on the vagus in the neck may slow the rate of ventricular contraction but the **P** waves continue to come through at *regular and rapid intervals*

The auricular contraction is no longer believed to pursue an abnormal circular path in the auricle When the normal rhythm is established as the result of the administration of digitalis or of quinidine the gentle undulations of the **P** waves of flutter are replaced by the sharper peaked waves of normal rhythm

ventricular premature systoles and with right bundle branch block. With right ventricular premature systoles and with left bundle branch block the deflections occur in the reverse direction Barker McLeod and Alexander (7) in 1930 introduced the new terminology which is the exact opposite of the old interpretation

Heart block Various degrees of impairment of conductivity of the heart muscle can be distinguished by the electrocardiograph

Slight Prolongation of the P-R interval to more than 0 2 second

Occasional Dropped Beats The P-R interval gradually becomes longer until finally the ventricular complex fails to follow its preceding auricular complex The P waves are quite regular but the ventricular complexes occur at different intervals and the P and T waves may at times fuse

Regular Dropped Beats Every fourth third or second ventricular contraction may be omitted constituting a 4 3 3 2 or 2 1 heart block Other varieties include 8 7 7 6 6 5 5 4 3 1 4 1 *and* 5 1 *heart block The first figure in each couple* indicates the number of auricular and the second the number of ventricular beats

Complete Heart block The ventricle contracts at its own independent rate of about 30 beats a minute The P waves occur regularly and may be seen isolated on the electrocardiogram or fused with R or T waves The ventricular complex is of normal duration and configuration and so the impulses giving rise to it are supraventricular but as they do not originate in the auricle they *must be derived from the junctional tissue (see Fig 18)*



Fig 18 Complete Heart block

Bundle branch Block and *Intraventricular Block* have already been described (p 178) (see Figs 10 11 and 12)

Sino auricular Block (Tortoise Heart) Some of the impulses arising in the S-A node fail to reach the main mass of auricular muscle and occasional intermittences of the whole heart may

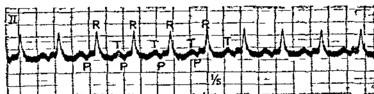


FIG Simple Paroxysmal Tachycardia showing inversion of the P wave

When the paroxysm ceases there is a pause which is immediately followed by the restoration of the normal rhythm

Auricular Flutter (see p 183)

Auricular Fibrillation (see p 184)

Nodal arising in the A–V node Here the auricle may contract first and the P–R interval be shortened and P inverted or the auricle and ventricle may contract simultaneously and the P and R be fused

Ventricular A rare condition characterised by a succession of ectopic ventricular systoles

Pulsus Alternans This condition which is revealed by a ✓ tracing from the arterial pulse is not always shown by the electro cardiograph Thus the R waves may be of equal height although the pulse shows marked alternation At times the R waves may show alternation but the large R waves may correspond with the small pulse waves and vice versa Occasionally the electrocardio gram shows alternation of the R waves although the pulse tracing is regular

Coronary Thrombosis Besides disturbances in rhythm coronary thrombosis often causes characteristic changes in the form of the ventricular complexes These changes were first observed by Smith (9) after experimental coronary occlusion in dogs and later by Herrick (10) and Pardee (11) in man Rothschild and his co workers (12) and Parkinson and Bedford (13) have demon strated that serial records at intervals after coronary throm bosis show that the electrocardiographic changes usually occur in a definite sequence corresponding with the patho logical events in progress in the myocardium

R–T Deviation Within a few hours up to a week of the onset the T waves take off from the RS waves above or below the

Auricular Fibrillation Here the P waves are absent QRS deflections occur at irregular intervals and are of varying height The ventricular complex is of normal form Oscillations of irregular size may be seen in diastole caused by the fibrillating auricle these may be fine or coarse They are usually best seen in Leads II and III They also occur during systole and may alter the shape of the T wave although the QRS deflections are not affected In rapidly beating hearts the oscillations are usually not well seen in diastole but their effect upon the T wave is generally noticeable (see Fig 21)

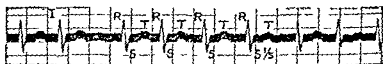


Fig 21 Auricular Fibrillation

The QRS deflections may occasionally be seen at regular intervals although the auricle is fibrillating and no P waves are present but oscillations due to fibrillation may be seen in diastole This occurs when the ventricle has adopted its independent rhythm owing to complete heart block

Ectopic beats may arise during fibrillation they are ventricular in origin They may occur regularly after each QRS deflection due to a supraventricular stimulus constituting the coupled beats This is usually due to excessive digitalis administration and is a sign for immediate discontinuance of the drug

Ventricular Fibrillation Although compatible with life in man if only of short duration (Callavardin and Berard (8)) it is the probable cause of sudden death from heart failure such as occurs under anæsthetics etc

Paroxysmal Tachycardia *Simple Paroxysmal Tachycardia (Auricular)* A succession of ectopic auricular contractions occurs arising at a new focus in the auricle This is shown in the electrocardiogram by a rapid regular rhythm of about 150 in which the ventricular complexes are normal The impulse has therefore a supraventricular origin The P wave is often inverted in Leads II and III, and usually modified in Lead I The auricular impulse is therefore ectopic in origin and if not inverted in Leads II and III it is usually smaller than normal (see Fig 22)

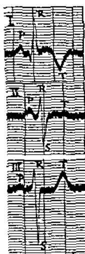


Fig 24 Electrocardiogram taken three weeks after coronary thrombosis showing the T1 type of curve (D E Bedford)

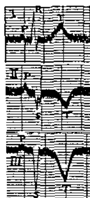


Fig 25 Electrocardiogram taken one month after coronary thrombosis showing the T3 type of curve (D E Bedford)

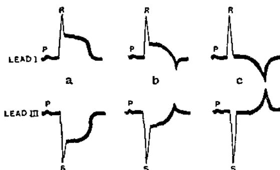


Fig 26 Diagram illustrating the changes in the ventricular complexes in Leads I and III after coronary thrombosis (a) R–T deviation (b) T waves becoming evident (c) R–T deviation disappeared The T waves point in the opposite direction to the R–T deviation in each lead (Parkinson and Bedford)

zero level so that RS is succeeded by a plateau shaped elevation or depression (see Fig 23) R–T deviation is most marked in Leads I and III and is in opposite directions in these two leads thus R–T elevation in Lead I is associated with R–T depression in Lead III and vice versa

T Wave Changes Within about a week the R–T segment returns to the iso electric level and T waves develop pointing away from the direction in which R–T was displaced in each lead Thus R–T elevation is succeeded by an inverted T and R–T depression by an upright T Inversion of T occurs either in Lead I or Lead III but not in both and often to a lesser degree in Lead II According to the direction of R–T displacement and consequently to the direction of the T waves in Leads I and III two types of curve are common

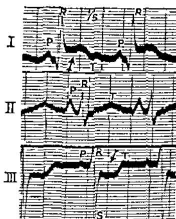


Fig 23 Electrocardiogram taken four days after coronary thrombosis showing plateau type of curve the R–T period is elevated in Lead I and depressed in Lead III as indicated by arrows (Parkinson and Bedford)

1 *T1 Type* T is deeply inverted in Lead I often slightly inverted or flattened in Lead II and upright in Lead III (see Fig 24)

2 *T3 Type* T is deeply inverted in Lead III often slightly inverted or flattened in Lead II and upright in Lead I (see Fig 25)

Changes in the direction of normal usually follow in the course of the first six months after the attack The inversion of T diminishes first in Lead II then in Lead I or Lead III eventually T may become upright in all leads but in some cases inversion of T persists almost unaltered Evidence is accumulating that the T1 type of curve corresponds with infarction of the anterior wall or apex of the left ventricle (occlusion of the left anterior descending coronary) and the T3 type of curve with infarction of the posterior wall of the left ventricle (occlusion of the right coronary (Barnes and Whitten (14))

Changes in QRS Notching and widening of QRS are quite

infarction Lead IV shows an abnormal curve in the majority of cases Thus using the right arm chest lead of Roth (Lead IV R) the typical findings are an elevation of the R–T segment with an inverted T wave The R wave is small or absent (see Fig 27)

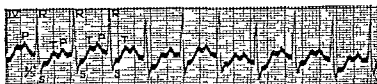


FIG 28 Lead IV R in T type of coronary thrombosis

The T wave may be huge over 10 mm In the T_3 type of electro cardiogram (see Fig 28) which is met with in acute posterior infarction the changes in the fourth lead are not so constant and not so characteristic as in the three conventional leads The R–T segment is depressed and the T wave is upright (see Fig 28)

When acute anterior and posterior infarctions occur simultan eously Wolferth and Wood (17) have shown that the R–T segment is raised in all the limb leads especially in Lead II In Lead IV R the R–T segment is also elevated Wood and Wolferth (18) suggest that the huge T waves (over 10 mm in amplitude) in Lead IV whether positive or negative indicate localised apical infarction

MULTIPLE CHEST LEADS

In 1938 (19) the American Heart Association endeavoured to standardise the use of multiple chest leads the chest electrode being placed on six different sites over the præcordium and on the left side of the chest These leads were not then adopted in Britain but the exact positions now recommended both by the American Heart Association and by the British Cardiac Society for chest leads are described later (see p 191) *With bipolar leads the distal* electrode which is paired with the chest electrode may be placed on the left leg the lead then being known as CF 1 to 6 according to the position of the chest lead or the distal electrode may be placed on the right arm, CR 1 to 6

frequent in curves after coronary thrombosis though not in any way special to this condition Low voltage of the *QRS* waves in all leads is also a common finding A large Q wave in Lead III has been noticed in many cases of coronary thrombosis and it is regarded as a suggestive sign of coronary disease though it occurs in the chronic stages and occasionally in other forms of heart disease or in displacement of the heart It is thought to indicate damage to the posterior part of the interventricular septum (Pardee (15) Fernichel and Kugell (16)) The reader s attention is also directed to the diagram (see Fig 26) which illustrates the electrocardiographic changes described above

Chest Leads in Coronary Disease

Wolferth and Wood (17) in Pennsylvania in 1932 introduced chest leads in the electrocardiographic study of coronary occlusion as there are ' silent areas in the heart where infarction may occur without producing a deviation of the S–T interval from the iso electric line in any of the three conventional leads The lead which is now commonly employed is IV R or CR IV The lead switch is turned to Lead I the left arm terminal is connected with the praecordial electrode and is placed on the extreme outer border of the apex beat as determined by palpation or auscultation The right arm electrode is placed on the right arm By this method both P and T waves are normally upright If the electrocardio graph cable has four leads the pectoral (PC) lead is attached to the praecordial electrode and the right arm lead attached to the right arm

The præcordial electrode should be circular 2 to 3 cm in diameter and made of German silver

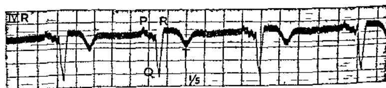


Fig 27 Lead IV R in T_1 type of coronary thrombosis

Results in Coronary Thrombosis In the T_1 type of electro cardiogram (see Fig 27) which corresponds with an acute anterior

away with the necessity of disconnecting one of the limb leads from the right arm left arm or left leg when taking the respective limb leads This disconnection is effected in the switch so that it is only necessary to turn the switch to the appropriate position as labelled in order to obtain all the leads required The positions on the unipolar lead switch we use are labelled S V aVr aVl and aVf With the unipolar lead switch turned to S the three standard limb leads can be taken by placing the patient switch on the electrocardiograph to position I II and III as for taking the ordinary bipolar limb leads By turning the patient switch back *to the Lead I position and the unipolar lead switch to position V* the 6 or more unipolar chest leads are taken by moving the chest electrode to the appropriate position By turning the unipolar switch to positions aVr aVl and aVf the unipolar limb leads are taken

The standard positions for the chest leads have been laid down by the Committee of the British Cardiac Society (22) in 1949 and they are as follows —

V1 right margin of sternum in the fourth intercostal space
V2 left margin of sternum at the same level
V4 mid clavicular line in the fifth left intercostal space
V3 a point midway between 2 and 4
V5 left anterior axillary line at the same level as V4
V6 left mid axillary line at the same level as V4
V7 left posterior axillary line at the same level
V8 *below the angle of the left scapula at the same level*
VE may be used the electrode being placed over the tip of the xiphoid cartilage

The corresponding V positions to the right are shown by the letter R *e g* V3R

If there is hypertrophy of the heart with a forcible impulse visible in the sixth space V4 must not be placed over the apex beat but in the position described above otherwise no information concerning rotation of the heart can be obtained

As the voltages of V leads are often high half standardisation may be used (1 mV = 5 mm) If this reduction is used it should be employed uniformly in all chest leads and indicated N/2

The electrode jelly must be carefully applied when recording the chest leads as positions 2 3 and 4 are often very close together

Unipolar Leads

In 1934 Wilson and his colleagues (20) introduced the unipolar leads These are known as V leads In this method the distal electrodes on the right arm left arm and left leg are connected each through a resistance of 5 000 ohms with a central terminal The chest electrode is then paired with the central terminal in order to take the chest leads which are known as V1 to V8 according to the position of the chest electrode

With bipolar leads the differences in potential between the two paired electrodes are recorded and the actual potential at either electrode is not indicated

With unipolar electrodes we have an exploring electrode placed either on the chest for the chest leads or on one limb for the limb leads and an indifferent electrode resulting from connecting the three limb electrodes to a central terminal The potential of the indifferent electrode may for clinical purposes be taken as zero although it can never actually become zero otherwise no current would flow Wilson has shown that the voltage at the so called indifferent electrode never rises above 0 3 mV With bipolar electrodes there are therefore two variables of potential whereas with the unipolar electrodes there is only one variable that of the exploring electrode

It was later noted that with the three distal electrodes connected with a central terminal in the majority of cases it made no difference in the electrocardiogram if the resistances were omitted from the circuit and in 1942 Goldberger (21) constructed a unipolar indifferent electrode which could be made in a few minutes at a cost of less than 10 cents Goldberger further found that he could augment the galvanometric deflection by 50% in the limb leads if the indifferent electrode was left off the limb from which the tracing was made These augmented limb leads are now generally employed and were known as aVR aVL and aVF a stands for augmented V for voltage indicating the unipolar lead and R L and F for right arm left arm and left leg respectively As augmented limb leads are now always employed the a is usually omitted

In order to take the unipolar leads whether chest or limb leads the switch on the electrocardiograph is placed in Lead I position Electrocardiographs are now supplied with a switch which does

away with the necessity of disconnecting one of the limb leads from the right arm left arm or left leg when taking the respective limb leads This disconnection is effected in the switch so that it is only necessary to turn the switch to the appropriate position as labelled in order to obtain all the leads required The positions on the unipolar lead switch we use are labelled S V aVr aVl and aVf With the unipolar lead switch turned to S the three standard limb leads can be taken by placing the patient switch on the electrocardiograph to position I II and III as for taking the ordinary bipolar limb leads By turning the patient switch back *to the Lead I position and the unipolar lead switch to position V* the 6 or more unipolar chest leads are taken by moving the chest electrode to the appropriate position By turning the unipolar switch to positions aVr aVl and aVf the unipolar limb leads are taken

The standard positions for the chest leads have been laid down by the Committee of the British Cardiac Society (22) in 1949 and they are as follows —

- V1 right margin of sternum in the fourth intercostal space
- V2 left margin of sternum at the same level
- V4 mid clavicular line in the fifth left intercostal space
- V3 a point midway between 2 and 4
- V5 left anterior axillary line at the same level as V4
- V6 left mid axillary line at the same level as V4
- V7 left posterior axillary line at the same level
- V8 below the angle of the left scapula at the same level
- VE may be used the electrode being placed over the tip of the xiphoid cartilage

The corresponding V positions to the right are shown by the letter R *e g* V3R

If there is hypertrophy of the heart with a forcible impulse visible in the sixth space V4 must not be placed over the apex beat but in the position described above otherwise no information concerning rotation of the heart can be obtained

As the voltages of V leads are often high half standardisation may be used (1 mV = 5 mm) If this reduction is used it should be employed uniformly in all chest leads and indicated N/2

The electrode jelly must be carefully applied when recording the chest leads as positions 2 3 and 4 are often very close together

The jelly in each lead must be kept separate otherwise if there is contact of the jelly the tracings will be distorted

There is a correlation between the standard limb leads and the unipolar limb leads for Lead VL = $\frac{I - III}{2}$ Lead VR = $-\frac{I + II}{2}$ and Lead VF = $\frac{II + III}{2}$

If the position of the heart in the thorax is considered it will be appreciated that the chest lead in position 1 and 2 is over the right side of the heart 3 is over the septal region 4 is over the septum or the left ventricle and 5 and 6 are over the left side of the heart With an extensive anterior infarction of the heart changes may be seen in all the chest leads 1 to 6 with an anterolateral infarct changes may occur in leads 4 to 6 and with an anteroseptal infarct in leads 1 to 3 The British Cardiac Society Committee state that " the full electrical exploration of the heart involves the taking of at least twelve leads and sometimes more The twelve basic leads are the three standard limb leads the three unipolar limb leads and six unipolar præcordial V leads For ordinary routine testing the following are recommended V1 and V2 or V4 and V5 or V5 and V6 according to the size of the heart together with VL and VF The right ventricular leads V1 and V2 show best some myocardial infarcts right bundle branch block right ventricular hypertrophy massive pulmonary embolism and auricular arrhythmias The left ventricular leads V4 to 6 are used to demonstrate some myocardial infarcts left ventricular hypertrophy and left bundle branch block Myocardial ischæmia is shown by V1 to 6 VL and VF For practically all purposes the leads taken may be reduced to the three standard leads with VF V4 and V6

Basic Normal V (Unipolar) Lead Patterns

Goldberger (23) described five basic ventricular unipolar lead patterns depending upon the surface of the heart the lead faces Further if the pattern shown in a limb lead is similar to that obtained from a chest lead it is presumed that the limb faces the surface of the heart from which the corresponding chest tracing is obtained

Electrocardiographically the ventricular muscle can be divided into three groups the interventricular septum, and the right and

left ventricles A transverse section through the chest at the level of the 4th costal cartilage shows that the right ventricle faces the anterior chest wall only a small portion of the right ventricle being found to the right of the sternum The greater part of the left ventricle lies over the diaphragm but in health these relationships vary according to the position of the heart The posterior wall of the heart is composed of the left auricle the left ventricle faces to the left side of the chest the right auricle to the right side of the chest and the interventricular septum runs obliquely from behind forward to the left its right ventricular surface facing anteriorly *and to the right its left ventricular surface facing posteriorly and* to the left

The unipolar leads whether chest or limb record alterations in potential produced by electrical changes in the ventricular muscle groups

With the heart in the usual position the chest leads V1 2 and 3 face the right ventricle and the right side of the septum V4 5 and 6 face the left ventricle and left side of the septum The limb lead VR faces the cavity of the right ventricle The limb leads VL and VF are much influenced by changes in the position of the heart

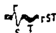
When a unipolar lead faces the epicardial surface of the left ventricle the ventricular complex consists of a small q wave followed by a large R and T is usually upright R T q qRT When the lead faces the left ventricle near the septum the q wave is absent but the R wave is large The initial small q wave is due to the electrical impulse spreading through the septum from the left to the right and so travelling away from the lead As soon as the stimulus spreads outwards through the left ventricle the R wave results The impulse is simultaneously spreading through the right ventricle which would give a downward deflection but this potential is too small to affect the R wave appreciably This pattern can be expected with V5 and V6 occasionally with V2 3 4 and with VL and VF

When a unipolar lead faces the epicardial surface of the right ventricle the ventricular complex consists of a small r and a large S or a large R and a large S T may be up or down r T S rST

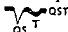
o R T S RST

Thus as the chest lead is moved from the V1 and V2 positions to the V5 and V6 positions the S wave disappears and the R wave increases in amplitude up to the V4 position and then diminishes again The point at which R = S is called the *transitional point* and indicates the line of the interventricular septum In some cases the transitional point is situated farther to the right than is shown in V1 or farther to the left than is shown in V6 There is usually a small q wave in V6 The T wave is often inverted in V1 and it may be inverted in V2 if the heart is vertical In children with normal hearts the T wave may be inverted in V3

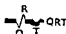
When a unipolar lead faces the cavity of the right ventricle as in VR the pattern of the tracing is rS with a downward T The r wave is due to spread of the impulse through the septum towards the right arm lead the S wave results from the stimulus spreading through the right and left ventricles away from the lead



When a unipolar lead faces the cavity of the left ventricle as in VL or VR or in a lead taken from the upper back of the chest the pattern is QS with a downward T The Q wave is due to the stimulus spreading through the septum away from the lead the S is due to the stimulus spreading through the ventricles still away from the lead



When a unipolar lead faces the back of the heart with the electrode placed in the interscapular region it faces the cavity of the left ventricle through the cavity of the left auricle and a portion of the epicardial surface of the left ventricle The pattern is QR followed by a downward T The Q is due to the lead facing the cavity of the left ventricle the R is caused by the stimulus passing out to the posterior wall of the left ventricle It will be seen that when the main QRS deflection is upward T also tends to be upward and vice versa A small round or U wave may occasionally be seen between the T and P waves



Axis Deviation

The unipolar leads enable a distinction to be made between deviation of the axis of the heart and preponderance of the ven

tricles Normally the cardiac axis runs obliquely downwards forwards and to the left The three standard leads six V leads and three unipolar limb leads with the heart in the normal semi vertical position are shown in Fig 29 The R wave in VF is tall and the deflections of VF resemble those of the leads from the left of the præcordium such as V6 When rotation occurs about a horizontal antero posterior axis it may be in a clockwise direction towards the vertical or in an anti clockwise direction towards the horizontal The heart may also rotate along its longitudinal axis If it rotates in a clock wise direction as looked at from below the right ventricle becomes more anterior and the left ventricle more posterior Rotation of the heart around its trans verse axis will bring the apex of the heart either nearer to or farther away from the anterior chest wall according to the direction of rotation

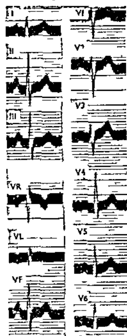


Fig 29 Semi vertical heart The heart is semi vertical because although there is a tall R wave in Lead VF the complexes of VL are small

Rotation around the horizontal antero posterior axis Six positions have been described vertical semi vertical inter mediate semi horizontal horizontal and indeterminate

With the vertical position the pattern of the ventricular complex in VL resembles that of V1 and V2 *i e* the right ven tricular pattern The ventricular complex of VF resembles that of V5 and V6 *i e* the left ventricular pattern (see Fig 30) When the heart lies in the horizontal position the ventricular complex of VL resembles that of V5 and V6 and the ventricular complex of VF resembles that of V1 and V2 (see Fig 31)

Rotation around the longitudinal axis In recording the chest leads of a normal heart the transitional point indicating the position of the interventricular septum is as described above between V3 and V4 leads If the heart rotates around the longi

tudinal axis in a clockwise direction the transitional point shifts to the left as between V5 and V6 The VL lead may face the right ventricular cavity when it will show a RS followed by a downward T wave (see Fig 32) If the rotation is in an anti

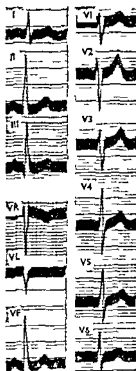


Fig 30 Vertical heart The heart is seen to be in the vertical position electrically because Lead VF shows a tall R wave and Lead VI a deep S wave



Fig 31 Horizontal heart The heart is horizontal because the R wave is tall in VL and the S wave is deep in VF Also the form of VL resembles that of V6

clockwise direction the transitional point shifts to the right between V1 and V2 or even farther to the right (see Fig 33)

Rotation around the transverse axis It is considered by some that the heart can rotate about a transverse axis and that this may affect the limb lead records although it does not appreciably affect those of the chest leads If the apex rotates backward

VF may face the right ventricle instead of the left ventricle as it usually does The left leg lead will now show a RS instead of the normal QR deflection

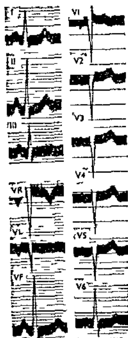


Fig 3 Considerable amount of clockwise rotation around the longitudinal axis The T P lies between V5 and V6 The heart is vertical

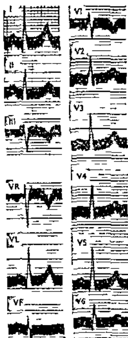


Fig 33 Considerable amount of anti clockwise rotation The T P is seen to lie between V1 and V2

Ventricular Hypertrophy

The diagnosis of hypertrophy of the ventricles from electro cardiograms still remains somewhat uncertain The curves obtained in the standard limb leads which were originally taken to indicate right or left ventricular preponderance were found at autopsy to be misleading in some cases The term axis deviation was then

used instead of ventricular preponderance for it was shown that with deep inspiration a tracing which was characteristic of left ventricular preponderance might become normal In long standing cases of ventricular hypertrophy changes occur in the standard limb lead curves the S–T segment is displaced in the opposite direction to the main initial deflection and the T wave also points away from the QRS Thus with advanced left ventricular preponderance RS–T is depressed and T is inverted in Lead I and RS–T elevated and T upright in Lead III In advanced right ventricular preponderance the reverse changes occur elevation of RS–T and upright T in Lead I and depressed RS–T and inverted T in Lead III These curves have been said to indicate *ventricular strain* although the term is not universally accepted Curves of this type probably indicate ischæmia of the right or left ventricle but not infarction

The chest unipolar leads are helpful in diagnosing right or left ventricular hypertrophy The chief electrocardiographic changes due to ventricular hypertrophy are high voltage QRS waves widening of the QRS so that it may resemble the complex characteristic of bundle branch block and delay in the intrinsic deflection in the leads over the hypertrophied ventricle The term intrinsic deflection which was introduced by Lewis and Rothschild is used to denote the moment when the impulse spreading outwards through the ventricular wall reaches the sub epicardial muscle It begins at the peak of the R wave and ends either at the isoelectric level or at the nadir of the S wave However in clinical cardiography it is usually measured from the beginning of the deflection to the peak of the upward deflection In human cardiography it should more properly be called the intrinsicoid deflection as the exploring electrode even in chest leads is comparatively far from the surface of the heart The R waves are small and the S waves deep in the tracings taken from the leads over the ventricle which is not hypertrophied Further in left ventricular hypertrophy the transitional point may be displaced to the right and in right ventricular hypertrophy to the left The electrocardiogram of left ventricular hypertrophy is shown in *Fig 34* Tall R waves and inverted T waves are seen in V5 and V6 with depression of the RS–T interval The T wave is inverted in VF In right ventricular hypertrophy as shown in Fig 35 the R waves

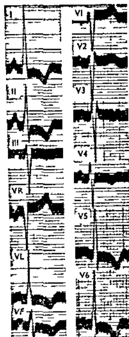


Fig 34 Left ventricular hypertrophy The R waves in V1 and V2 are even smaller than normal but the S waves are deeper than normal In V5 and V6 the peak of R is abnormally late R is too tall the RS T segment is depressed and T is inverted

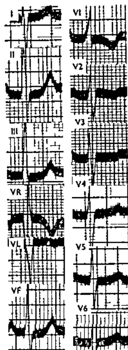


Fig 35 Right ventricular hypertrophy In V1 V2 and V3 R is abnormally large and its peak is late QRS is a little widened and T is inverted

are tall and T inverted in V1 V2 and V3 QRS is a little wide and the T wave is inverted in VR Depression of the RS–T segment and inverted T waves in the chest leads facing the epicardial surface of the affected ventricle with normal QRS complexes is said to indicate ischæmia It can be shown with the combination of the chest and limb leads that right axis deviation may be present with left ventricular hypertrophy

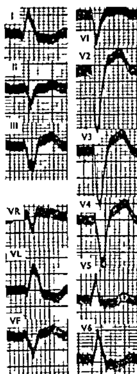


Fig 36 Complete left bundle branch block QRS is at least of 0·12 sec duration In V1 and V2 R is small or absent and a deep broad S is present In V5 and V6 a broad R occurs which may be notched or bifid

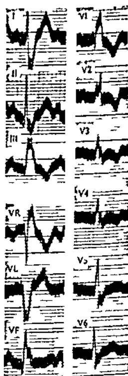


Fig 37 Complete right bundle branch block The QRS complex is prolonged to 0·12 sec or more V1 and V2 show a broad notched R wave which is preceded by a very small primary R and S wave In V5 and V6 the R wave is slender and followed by a broad S It is seen that the pattern of right bundle branch block can closely simulate that of right ventricular hypertrophy

Bundle Branch Block

The use of chest leads has confirmed the view that the common divergent or Type I bundle branch block as shown in the standard limb leads is due to left bundle branch block The characteristic features of the unipolar chest leads in bundle branch block are as follows —

Left Bundle Branch Block Leads V1 and V2 facing the right ventricle show r small or absent S deep and broad Leads V5 and V6 facing the left ventricle show R large broad and notched (see Fig 36) If a small q wave is present there is probably left ventricular hypertrophy and not bundle branch block

Right Bundle Branch Block Leads V1 and V2 show a wide **RSR** or **RSR S** with usually a downward T wave Leads V5 and V6 show a narrow **qR** wave followed by a wide S T is usually upright (see Fig 37) The pattern closely resembles that of right ventricular hypertrophy but in the latter the **qRs** pattern is normal in Leads V5 and V6

Goldberger (93) states that left bundle branch block may be transient or permanent As a transient condition it may occur for no apparent reason or following infarction congestive failure during acute infections or it may result from quinidine Permanent left bundle branch block because it is almost always due to organic heart disease usually shortens life Right bundle branch block may also be transient or permanent The transient variety may occur for no apparent reason or follow pulmonary embolism or myocardial infarction Permanent right bundle branch block is associated with rheumatic arteriosclerotic and hypertensive heart disease and coronary occlusion When myocardial infarction and pulmonary embolism are absent right bundle branch block is not of serious significance

Myocardial Infarction

When a portion of the wall of the heart is the site of an infarct it becomes electrically inert and an electrode placed opposite this area is described picturesquely as looking into the cavity of the ventricle through a window of inactive tissue The **Q** wave may be abnormal in depth and width signifying the presence of dead muscle tissue The electrode registers the negative voltage of the ventricular cavity The **R** wave is diminished in size over the damaged muscle Whether or not it is present depends on whether there is muscle on the surface of the infarct A tall **R** wave may occur at the edge of the infarct The **RS–T** segment is displaced above or below the iso electric level An electrode facing the injured surface will show a rise in the **RS–T** segment an electrode facing in the opposite direction will show a depressed **RS–T** segment

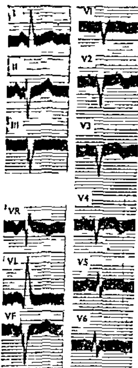


Fig 38 Strictly anterior infarct The infarct does not reveal itself in the standard leads V1 to V4 are chiefly affected

The T wave points in a direction opposite to that of the RS–T displacement

The unipolar chest leads enable the site of the infarct to be determined with some degree of accuracy The following types are described —

Anterior infarction The standard leads are often normal in this type of infarction V1 2 and 3 however show the displaced RS–T segment and inverted T waves (see Fig 38)

Antero septal infarction If in the above type of curves the QRS is widened and the Q waves are deep in V1 2 and 3 the infarct has probably spread into the septum with involvement of the right bundle branch

Antero lateral infarction V4 5 and 6 show the characteristic changes They are also seen in the standard Lead I and unipolar Lead VL (see Fig 39)

Extensive anterior infarction Changes are seen in all the chest leads and T is inverted in standard Lead I and in unipolar Lead VI

Posterior infarction The chest leads are often of no help in diagnosing posterior infarction although occasionally the T waves are abnormally high The important lead here is VF which shows elevation of the RS–T segment and sharply inverted T waves Lead III shows similar changes (see Fig 40) Similar changes in the standard Lead III may occur without infarction in which case VF is normal The unipolar lead VF will indicate whether a suspected Q_3T_3 in the standard Lead III indicates infarction or not or whether the pattern in Lead III is really S_3T_3 indicating a transverse lie of the heart When in addition to the changes in VF V6 or V5 and V6 show an abnormal RST the infarct may be described as postero lateral

Septal infarction Here the standard limb leads indicate a

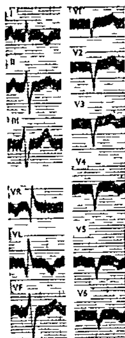


FIG 39 Antero-lateral infarct Maximal changes are seen in V4 to V6 but often all chest leads are involved T is inverted always in VL and I

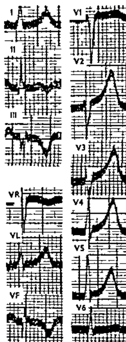


FIG 40 Posterior infarction The typical changes are seen in Leads VF and III Although the V chest leads are often disappointing they may show abnormally tall and symmetrical T waves as in this case

posterior infarction and the chest leads indicate an antero-septal infarction

Acute Pericarditis

When the epicardial surface of the heart is injured the electrode facing the injured surface will show elevation of the RS–T segment Thus both myocardial infarction if it involves the epicardial surface of the heart and pericarditis if that also does so will both show RS–T segment changes With infarction a pathological Q wave is usually also present but it is absent in pericarditis

If we consider three leads namely VL VF and an anterior chest

lead it will be seen that in anterior infarction the **RS–T** segment is elevated in **VL** and also in the chest lead but is depressed in **VF**. Conversely in posterior infarction the **RS–T** segment is depressed in **VL** and in the chest lead (although commonly it is not very greatly depressed in chest leads) and elevated in lead **VF**.

In generalised pericarditis leads facing both the front and back of the heart will show elevation of the **RS–T** segment. Therefore the **VL**, **VF** and chest leads all show elevation of the **RS–T** segment.

When pericarditis is complicating anterior infarction lead **VL** will show elevation of the **RS–T** segment both as a result of the infarct and of the pericarditis and a deep **Q** wave will be present in this lead. Lead **VF** will show elevation of the **RS–T** segment owing to the pericarditis at the back of the heart but the **Q** wave will be normal. The chest leads will show elevation of the **RS–T** segment owing to the presence of the infarct and the pericarditis and pathological **Q** waves will be present too.

When pericarditis complicates posterior infarction lead **VL** will show elevation of the **RS–T** segment owing to the pericarditis at the front of the heart but no pathological **Q** wave will be present. Lead **VF** will show elevation of the **RS–T** segment due both to the posterior infarct and the pericarditis at the back of the heart and a pathological **Q** wave will be present as a result of the infarct. The chest leads will show elevation of the **RS–T** segment owing to the pericarditis but no pathological **Q** wave will be present. Fig. **41** illustrates the five possibilities —

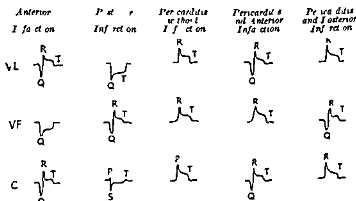


Fig. 41 Diagram showing electrocardiographic changes in Infarction and Pericarditis

Massive Pulmonary Embolism

In acute cor pulmonale associated with an extensive pulmonary embolus electrocardiographic changes may be found In the standard limb leads there is a deep S wave in Lead I and a Q and inverted T wave in Lead III together with an inverted T wave in the chest lead V1 These changes do not usually persist for any length of time although rarely they may be seen for several months Transient right bundle branch block may also be associated with pulmonary embolism

Conclusions The use of the unipolar leads has enabled a more exact localisation of myocardial infarcts to be made but this does not imply any alteration either in the treatment of the case nor in our views as to prognosis These leads have also allowed a more definite diagnosis to be made in cases of suspected posterior infarcts Bundle branch block the presence of ventricular hypertrophy and changes in the position of the heart can also be diagnosed more accurately The superiority of the unipolar over the bipolar chest leads is not universally accepted Thus Evans (24) states Anyone who customarily used CR leads need not change to the unipolar chest leads in the hope that they would gain additional information in the clinical diagnosis Evans says that CR_1 portrays P waves better than does V1 and so is more useful in arrhythmias Further the T wave is often inverted in healthy adults in V1 rarely so in CR1 as the R wave is better portrayed in CR1 than in V1 It will be admitted by all that the reading of the unipolar leads is often difficult and uncertain and the theoretical basis for their interpretation is at times unconvincing For more detailed information on unipolar leads the reader is referred to the book by Goldberger (23) and to articles by Hill (25) and Oram (26) on whose writings this section is largely based

Cardiac Catheterisation

By the use of what is known as the Fick principle physiologists have determined the cardiac output in dogs The method depends upon ascertaining the output of the right ventricle from the amount of blood passing through the lungs in a given unit of time The output of the right ventricle equals that of the left The volume of blood passing through the lungs can be calculated if the amount

lead it will be seen that in anterior infarction the **RS–T** segment is elevated in VL and also in the chest lead but is depressed in VF. Conversely in posterior infarction the **RS–T** segment is depressed *in VL and in the chest lead* (although commonly it is not very greatly depressed in chest leads) and elevated in lead VF.

In generalised *pericarditis* leads facing both the front and back of the heart will show elevation of the **RS–T** segment. Therefore the VL, VF and chest leads all show elevation of the **RS–T** segment.

When pericarditis is complicating anterior infarction lead VL will show elevation of the **RS–T** segment both as a result of the infarct and of the pericarditis and a deep **Q** wave will be present in this lead. Lead VF will show elevation of the **RS–T** segment owing to the pericarditis at the back of the heart but the **Q** wave will be normal. The chest leads will show elevation of the **RS–T** segment owing to the presence of the infarct and the pericarditis and pathological **Q** waves will be present too.

When pericarditis complicates posterior infarction lead VL will show elevation of the **RS–T** segment owing to the pericarditis at *the front of the heart* but no pathological **Q** wave will be present. Lead VF will show elevation of the **RS–T** segment due both to the posterior infarct and the pericarditis at the back of the heart and a pathological **Q** wave will be present as a result of the infarct. The chest leads will show elevation of the **RS–T** segment owing to the pericarditis but no pathological **Q** wave will be present. Fig 41 illustrates the five possibilities —

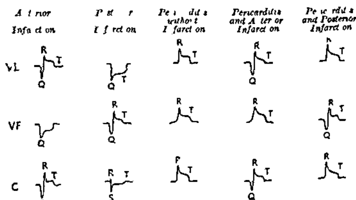


Fig 41 Diagram showing electrocardiographic changes in Infarction and Pericarditis

As the catheter passed through the subclavian vein I experienced an intense feeling of warmth behind the collar bone under the insertion of the sternomastoid muscle and at the same time through irritation of the branches of the vagus a dry cough I proved the position of the catheter by an X ray picture and I observed the progress of the catheter myself on the screen in a mirror held by the sister

A truly remarkable experiment !

Since then the method of cardiac catheterisation in man has become more and more extensively employed and reference will be made to some of the publications during the succeeding years In 1932 Grollman (28) employed catheterisation of the right auricle to determine the cardiac output in man Further advances were made in 1941 by Cournand and Ranges (29) They allowed the catheter to remain in the heart for an hour or longer They were thus enabled to determine the intra auricular pressure and by analysing the gas content of the auricular blood and of arterial blood they determined the cardiac output according to the Fick principle In 1944 McMichael and Sharpey Schafer (30) published their observations on the effect of posture venous pressure changes and the administration of atropine and adrenaline on the cardiac output in man as determined by cardiac catheterisation Their investigations were made on normal male volunteers and took place two hours after the midday meal A cardiac catheter was passed into the right auricle and the oxygen unsaturation of the right auricular blood was determined by a Haldane gas analysis apparatus The oxygen capacity was determined by hæmoglobinometry and with normal lungs arterial puncture was not performed It was assumed that arterial blood was 95 % saturated and in this way the arteriovenous oxygen difference was calculated The mean figure for the arteriovenous oxygen difference was in the region of 45 ml /litre The average resting cardiac output in the recumbent position was 5 3 litres a minute and in the standing position 4 litres a minute Raising the heart rate by the intravenous injection of 1 mg of atropine increased the cardiac output and caused a fall in the intra auricular pressure Intravenous injection of adrenaline increased the cardiac output in doses that did not accelerate the heart or raise the blood pressure Sharpey Schafer (31) in 1944 made observations by means of cardiac catheterisation on

of carbon dioxide given off or the amount of oxygen taken up is determined together with the arteriovenous difference of either of these gases The oxygen utilised or the carbon dioxide produced can be ascertained by spirometric and gas analysis methods The arterial blood is obtained by direct arterial puncture and the mixed arteriovenous blood is withdrawn either by puncture of the right ventricle or by passing a catheter down the external jugular vein into the right ventricle *The samples of blood are then analysed either for their O_2 or their CO_2 content. If the O_2 content is* determined the cardiac output is determined from the formula — Cardiac output *in litres per minute* = O_2 *consumption in ml per minute divided by the arteriovenous O_2 difference in ml per litre* Thus if the O_2 consumption is 250 ml/minute the arterial O_2 content 200 ml /litre and the venous O_2 content 150 ml /litre then the

$$\text{cardiac output} = \frac{250}{200-150} = 5 \text{ litres per minute}$$

Forssmann (27) in 1929 passed a ureteric catheter through a vein *in his arm into his heart His paper makes interesting reading* and the translation of the relevant section is as follows —

After successful experiments in passing a catheter on the cadaver *I undertook on myself the first experiment on living man First I submitted myself to an experiment by a colleague who* kindly placed himself for this purpose at my disposal and punctured with a thick needle my right antecubital vein I then passed as *in the experiment on the cadaver a well oiled ureteric catheter* size 4 Charrieres through the cannula in the vein The catheter was passed quite easily for a distance of 35 cm It then appeared to my *colleague* to be dangerous to go farther so the experiment was stopped *although I was quite undisturbed*

A week later *I undertook another experiment on my own As* a vein puncture with a thick needle on one's own body presents certain technical difficulties with a local anæsthetic I made a vein section in my *left antecubital fossa and passed a catheter* with little resistance *for its whole length of 65 cm* This length appeared to me after measuring the distance on the surface of the body to correspond with the distance from the left elbow to the heart All I felt during *the passage of the catheter was a slightly warm* sensation *such as results from an intravenous injection of calcium*

that the average difference between ventricular and auricular samples was for CO_2 0 42 vols % and for O_2 0 26 vols %

Work was now directed towards the investigation of congenital heart disease by means of cardiac catheterisation Brannon et al. (35) demonstrated that in cases of atrial septal defects oxygenated blood passes from the left to the right atrium In cases of hypertrophy of the right ventricle and prominent pulmonary arteries due to other causes cardiac catheterisation revealed no evidence of an atrial septal defect

Dexter et al (36) in 1946 showed that the cardiac catheter could be passed into the pulmonary artery and its branches In 5 cases of patent ductus arteriosus they found a higher oxygen content in blood drawn from the pulmonary artery than in blood taken from the right ventricle This was due to oxygenated blood passing from the aorta via the ductus to the pulmonary artery

Johnson et al (37) in 1947 introduced the method of heparinising the patient and not the saline This is referred to later under the section Variations in Technique

Howarth et al (38) investigated cases of congenital heart disease The magnitude of a left to right shunt through an auricular septal defect can be calculated from a comparison of the gas content of specimens removed from the superior and inferior venæ cavæ with that of specimens obtained from the right auricle and right ventricle Blood obtained from the right auricle and right ventricle contained more oxygen than vena caval blood if the shunt is from left to right A reduced arterial oxygen saturation when the lungs are functioning normally suggests right to left shunt In pulmonary hypertension with enlarged pulmonary arteries and a normal arterial oxygen saturation the oxygen content of the blood obtained by cardiac catheterisation showed no evidence of shunt but the mean right ventricular pressure was greatly increased

Holling and Zak (39) in 1950 published their observations on cardiac catheterisation of 70 cases of congenital heart disease which are referred to later

The Method

The requirements are as follows —

Cardiac Catheters These are of a modified ureteral type

the cardiac output in severe anæmias He found that the resting minute oxygen consumption was not reduced in chronic and post hæmorrhagic anæmia The cardiac output was increased up to 12 to 14 litres per minute which helped to maintain the oxygen supply There was also an increased removal of oxygen in the periphery and a reduced blood volume which caused a greater total hæmoglobin concentration McMichael and Sharpey Schafer (32) in 1944 used the method of cardiac catheterisation to determine the effect of digoxin on the cardiac output of man in normal persons and in patients suffering from cardiac failure In all cases there was a fall in the right auricular pressure It was found that if the initial cardiac output was normal or high the fall in auricular pressure was accompanied by a fall in cardiac output If however the cardiac output was low the fall in auricular pressure was accompanied by a rise in output There may therefore, be cardiac failure with high or with low output Digitalis lowers the venous pressure and the cardiac output in normal persons and in patients suffering from some types of anæmia and cor pulmonale and so they concluded that digitalis is unlikely to benefit the group of cases in which there is a high cardiac output This view however is not universally accepted In the low output cases of heart failure which includes most cases of congestive failure lowering of the venous pressure by digitalis and other methods is often followed by a rise of cardiac output The authors also concluded that as mechanical lowering of the right auricular pressure produces effects on cardiac output similar in degree to those produced by digoxin the major beneficial effect of digoxin results from a peripheral action on venous pressure and not from a tonic effect upon the heart The slowing of the cardiac rate by digoxin in cases of auricular fibrillation did not appear to have a beneficial effect on cardiac output

Cournand (33) in 1945 described his modified technique for sampling blood from the right auricle and the femoral artery and Cournand et al (34) in the same year published further observations on measuring the cardiac output in man by catheterisation of the right auricle or ventricle The catheter was left in the heart for up to twenty four hours and a special needle was devised for cannulisation of the femoral artery the needle was strapped in position and a lock syringe attached for sampling They found

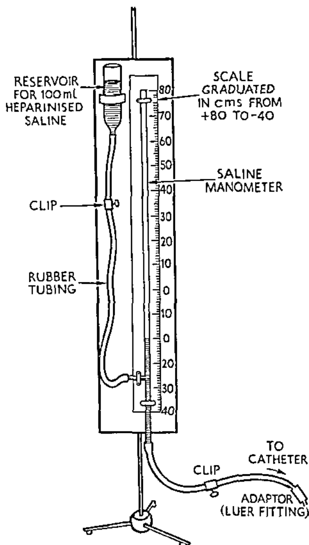


FIG 4 Reservoir and Manometer for Cardiac Catheterisation

made of nylon with a smooth unwettable plastic covering flexible and radio opaque The catheters are obtainable from the U S Catheter and Instrument Co Glens Falls N Y They are 100 cm or more in length and various sizes should be available such as 6 7 8 and 9 For an infant a No 6 will be required while a No 8 is used for the average adult A short distance from the tip of the catheter there is a slight bend or angulation which enables the direction to be changed by rotation of the external end A Luer fitting metal adaptor is fitted to the external end The catheters are sterilised by means of formalin vapour

A Saline Manometer This is used to record the mean pressures in the chambers of the heart and is sterilised by steam It is mounted on a stand which rests on the floor adjacent to the table on which the patient lies It can be adjusted in the vertical plane so that the zero mark can be set level with the sternal angle Alternatively a metre rule with spirit level attached can be used to determine the zero level on the manometer in which case the manometer is not moved up and down to the level of the sternal angle The metre rule is placed horizontally from the angle of the sternum to the manometer and the marking on the manometer which corresponds with the zero reading is noted To the lower end of the manometer is connected a sterile rubber tubing with an adjustable clip attached The distal end of the rubber tubing is connected as required with the Luer fitting adaptor of the catheter A short distance above the lower end of the manometer is a side tube to which is connected by a sterile rubber tube fitted with an adjustable clip a saline reservoir held by clips to the top of the manometer stand The reservoir previously sterilised by boiling is nearly filled with sterile physiologic saline The manometer is filled almost to the top from the reservoir before use and it can be replenished by releasing the clip on the rubber tubing connecting the two (see Fig 42)

Sterile Screw capped Bottles These contain a few ml of sterile paraffin and one drop of heparin solution These bottles are for the collection of blood samples for gas analysis

Cutting Down set for skin incision and isolation of vein

Sphygmomanometer or tourniquet

Hypodermic syringes 1 ml 2 ml 5 ml and 20 ml

Hypodermic needles Nos 20 17 15 and 1

taken to avoid exposure of the hands since it is not possible to wear leaden gloves. X-ray screening should be limited to about 25% of the erythema dose, which means about twenty minutes in all. Wood (40) states that the screening time should be limited to thirty minutes at 1 mA or twenty minutes at 2 mA or fifteen minutes at 3 mA. The same operator should not regularly perform more than two catheterisations a week. It is desirable but not essential to have facilities for taking films to check the position of the catheter subsequently. The patient should be made as comfortable mentally and physically as possible before the operation starts and its nature should be explained to an adult. The cardiac output may be increased by anxiety without a corresponding increase in the oxygen consumption. This will decrease the arteriovenous difference. A vein in either arm may be used for the catheterisation but the left is preferable as the course of the veins to the right auricle is a more gradual curve than on the right side and so a hold up is less likely to occur. Whichever arm is selected the cephalic vein should not be used as it joins the axillary vein almost at a right angle which causes difficulty in passing the catheter beyond this point. The medial antecubital vein is best suited for the operation and the point of insertion of the catheter should be a short distance distal to where two veins join. This will ensure a continuous flow of blood along the catheterised vein both during and after the operation and will minimise the risks of thrombosis.

The selected arm is abducted to about 45° and bound lightly to a well padded arm splint. The sphygmomanometer cuff should be in place and all but the antecubital fossa covered with sterile mackintoshes and towels. The exposed skin is cleaned with ether and the veins rendered prominent by inflation of the sphygmomanometer cuff. The skin and subcutaneous tissues around the site of the proposed incision and the perivenous tissues are now infiltrated with 2% procaine solution. Adequate intradermal anæsthesia with the formation of a wheal at least the size of a half crown is important to prevent the subsequent occurrence of venospasm. An incision about 1 cm long is then made through the skin in line with the vein and the vein isolated by blunt dissection. Two fine catgut ligatures are passed round the vein and the distal one is tied off but not cut. The upper ligature is

Procaine hydrochloride solution 2%

Serum needles 2½ inch 17 gauge

Ten 10 ml all glass syringes Luer fitting The syringes are sterilised by boiling in water and then lubricated internally with sterile paraffin

Ten blood sampling bottles as described above

One ampoule of heparin containing 50 mg

Catgut No 0000

Sterile normal saline

Ether for skin preparation

Sterile gowns caps and masks

Sterile mackintoshes towels towel clips and dressings

Sterile bowls and gallipots

On an emergency trolley should be placed an oxygen apparatus and B L B mask A mouth gag Tongue forceps Brandy coramine adrenaline and amyl nitrite Two fine needles (4 inches long) for emergency cardiac puncture

Preparation of the Patient The patient's sensitivity to quinidine sulphate should be tested two days beforehand by giving 3 gr If he shows no idiosyncrasy 5 gr of quinidine sulphate and 3 gr of Amytal are given by mouth one hour before the operation The quinidine is given to try and prevent cardiac irregularities from developing during the catheterisation It is a wise precaution to give 100 000 units of penicillin intramuscularly twice a day on the day of the operation and to continue with this dosage for the next two days Phenobarbitone 1 to 3 gr by mouth may be given to an adult and to children over ten years of age two hours before the operation Children under ten years of age require a general anæsthetic and rectal Pentothal 1 g per 50 lb of body weight is satisfactory If necessary this dose can be supplemented later by injection of more Pentothal through the cardiac catheter Inhalation anæsthesia is unsuitable as it interferes with the blood gas analysis

The Operation The operation is carried out in the X ray department with the patient lying horizontal on a table covered with a rubber mattress The table is provided with a horizontal fluoroscopic screen controlled by a foot switch In addition to observing rigid surgical asepsis operators must be well protected from scatter by means of lead aprons Particular care should be

is prolonged Further Warren (41) states that samples of blood from the proximal pulmonary artery appear to be most representative of mixed venous blood samples from the ventricle or atrium are more variable

Recording the Intracardiac Pressure With the tip of the catheter in the pulmonary artery the manometer is connected by the rubber tubing to the outer end of the catheter in place of the syringe care having been taken to exclude all air from the rubber tubing by allowing the heparinised saline in the reservoir to run freely through it and then clipping the tubing between the manometer and catheter and the reservoir and manometer in this order The clip between the manometer and catheter is now released and the saline level in the manometer will gradually fall until the pulmonary artery pressure is reached It will then remain stationary or oscillate slightly The reading is noted the clip between the manometer and the catheter closed and that between the reservoir and manometer opened so that the manometer is again filled

Removal of Blood Samples The rubber tube from the manometer is disconnected from the catheter and 3 or 4 ml of blood are drawn back into a saline filled syringe These are discarded as they will be diluted with the saline in the catheter A sterile 10 ml syringe previously lubricated with sterile paraffin and containing about 1 ml of paraffin free from air bubbles is now attached to the catheter and 5 to 10 ml of undiluted blood are withdrawn The syringe is detached and the blood is injected through a serum needle under the liquid paraffin in one of the specimen bottles and stirred to mix with the heparin to prevent clotting In order to prevent air embolism the hilt of the catheter must be kept below the level of the right auricle when removing samples of blood The outer end of the catheter must never be left open so that when a syringe containing a blood sample is detached either a fresh syringe filled with saline or the manometer tubing should be immediately attached in its place A little heparinised saline is now allowed to run through the catheter to displace the blood contents

The catheter is now gradually withdrawn further pressure readings and blood samples being taken from the upper part of the right ventricle the lower part of the right ventricle the right

not tied but its ends *are secured with Spencer Wells forceps* This ligature is used to apply gentle traction on the vein to steady it while it is opened and to control hæmorrhage To the external end of the catheter which has been well rinsed inside and out with sterile normal saline is attached a 10 ml all glass syringe charged with sterile normal saline containing 25 mg of heparin All air is expelled from the catheter by depressing the plunger of the syringe *and both are laid on a sterile towel close to the operator s right* hand *The other 25 mg of heparin are added to the saline in the* reservoir connected to the manometer and the latter is filled to near the top with this solution

Passing the Catheter The segment of exposed vein is now raised by traction on the proximal loop of catgut and a small fish mouth incision is made in its wall with pointed scissors The tip of the catheter is immediately inserted into the vein and gently *pushed up its lumen for several centimetres The heparin solution* is then injected via the catheter and the syringe is left attached to its outer end Further progress of the catheter is watched on the fluoroscopic screen with short exposures at frequent intervals Force must be avoided and considerable patience is sometimes necessary Entry of the catheter into the neck veins is prevented by turning the patient s head to the side of the operator and if *obstruction occurs at the thoracic inlet slight movement of the arm or shoulder* or a deep inspiration by the patient will usually permit it to pass easily Should the catheter take a wrong direction it must be withdrawn some distance and another attempt made after rotation of its external end Once it has reached the superior vena cava it passes readily down into the right auricle Manipulation of the outer end *will direct it through the tricuspid valve into the cavity of the right ventricle Premature systoles are commonly* observed when the tip is in the vicinity of the tricuspid ring and delay at this point should be avoided if possible Having entered the right ventricle the catheter can be persuaded to turn upwards into the outflow tract thence through the pulmonary valve into the main pulmonary artery and *if further advanced it will enter one or* other pulmonary artery *It is wise to pass the catheter* as far as the pulmonary artery before taking the first blood sample and pressure reading for venospasm may occur and prevent further forward progress of the catheter especially if the operation time

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The catheter is now gradually withdrawn further pressure readings and blood samples being taken from the upper part of the right ventricle the lower part of the right ventricle the right

and auricle and the superior vena cava The normal mean pressures referred to the sternal angle level are — Pulmonary artery 18 to 20 cm saline (10 to 15 mm Hg) Right ventricle 11 to 16 cm saline (8 to 12 mm Hg) Right auricle —3 to +3 cm saline (—2 to +2 mm Hg)

The catheter is then completely withdrawn and the proximal catgut ligature tied to arrest bleeding A skin suture may not be necessary but should be available After the operation movements of the arm should be encouraged to minimise risk of thrombosis Immediately after withdrawal the catheter must be well washed through with water under pressure for several minutes then hydrogen peroxide (10 vols) is trickled slowly through until bubbling ceases and it is finally washed again with tap water The blood samples should be analysed immediately and not allowed to stand overnight Arterial puncture is only required when the cardiac output is being determined and then some authorities do not perform it their calculations being based on the assumption that the arterial blood is 95 % saturated with oxygen In cases in which there is a shunt between the two sides of the heart the cardiac output cannot be determined by the Fick principle If arterial blood is required direct puncture is employed It is thought by Cournand that the brachial rather than the femoral artery should be punctured The tissues around the artery are first infiltrated with - procaine solution For serial observations an indwelling needle is employed A special Cournand needle is used and is inserted with the sharp open bore stilet in place When the artery is pierced a small drop of blood appears through the fine bore The sharp stilet is now removed when the blood spurts out The sharp stilet is now replaced by a blunt one and the needle is threaded up the artery a short distance and is then strapped in position When the stilet is removed for blood sampling it should stand in peroxide solution to prevent clotting when it is replaced The blood samples are removed as is done for the intra cardiac samples When the cardiac output is being determined the oxygen consumption is determined at the end of the operation using the Benedict Roth apparatus

Variations in Technique The catheter may be sterilised by boiling for ten minutes it is then left to cool in the sterile towel until it becomes sufficiently rigid for use

Some workers employ a three way stopcock to connect the fluid in the reservoir the manometer and the sampling syringe with the catheter A Tycos manometer may be used instead of a saline one In this case a few ml of saline solution are introduced into the tubing in the part adjacent to the catheter attachment to minimise the risk of air embolism There is thus an air lock between the manometer and the saline in the manometer tubing Johnson et al (37) take the zero level as one third of the distance from the sternum to the back at the level of the third costal cartilage The saline manometer readings average 11 mm less than those recorded by the Tycos instrument The saline manometer has the disadvantage of reacting slowly especially when the catheter is full of blood The Tycos manometer is brisk in its response Electrical recording devices are now being more commonly employed These give pulse tracings from which systolic diastolic and mean pressures can be recorded and the form of the pulse wave studied In young children the catheter may be introduced into the saphenous vein and passed up the inferior vena cava

When a three way stopcock is used the tap is placed so that the lumen of the catheter communicates with the fluid in the reservoir and as the catheter is introduced into the vein a slow flow of fluid is maintained through the catheter at a rate of 20 to 30 drops a minute As soon as the catheter is inside the heart shadow the saline flow is adjusted to 5 or 10 drops a minute

Silk ligatures may be employed instead of catgut ones At the end of the operation both ligatures are removed and bleeding is controlled by pressure When the arterial needle is removed firm pressure must be maintained over the artery for at least five minutes and the pulses below carefully checked

Some workers such as Johnson et al (37) prefer to heparinise the patient instead of the saline In this case a heparin curve must be done the day before the operation from which the necessary dose can be worked out to maintain the clotting time during the operation and for twenty four hours afterwards at ten to twenty minutes (normal three to seven minutes) by the capillary method

Cournand et al (42) advise that a direct vision electrocardiograph should be used during catheterisation to record the cardiac rhythm and enable abnormalities to be detected immediately

Complications Warren (41) says that several thousand human cardiac catheterisations have been done in various laboratories with no record of serious untoward effect There are however certain risks attached to the procedure

General Reactions Fainting sweating nausea or vomiting may occur during catheterisation Rigors are due to pyrogens in the catheter which has not been properly cleaned

Veno spasm This is due according to Wood (40) to the skin not being adequately anæsthetised to the catheter not being washed free from disinfectant or to the catheter being too large

Air Embolism Strict precautions are essential to prevent air entering the catheter for in cases of congenital heart disease with right to left blood shunts air emboli could pass into the systemic circulation This has very rarely occurred the air bubbles passing to the brain producing transient neurological signs

Thrombosis This may occur either in the veins or in the heart If the saphenous vein is used it should be tied off as close to the femoral vein as possible at the end of the operation Slight local thrombophlebitis at the site of the introduction of the catheter into the arm vein has been from time to time recorded Johnson et al (37) record the case of a markedly cyanotic infant weight 19 lbs A catheter was introduced into the right saphenous vein near the femoral junction and passed easily into the right auricle The infant was heparinised during the operation A month later death occurred when a clinically unsuspected well organised thrombus was found occluding the inferior vena cava both common iliac veins and the right renal vein and a large thrombus was attached to the right auricular wall at the base of one leaflet of the tricuspid valve Holling and Zak (39) found evidence either of systemic or pulmonary thrombosis or emboli in 6 of their 70 cases of congenital heart disease within fourteen days of catheterisation and one case died

Disturbances of Cardiac Rhythm Cournand et al (42) consider these are the most important complications in cases of congenital heart disease They include premature contractions runs of ventricular tachycardia ventricular fibrillation and auricular tachycardia If cardiac catheterisation is done under electrocardiographic control the disturbance of rhythm can be

recognised early and the catheter withdrawn These workers record sudden death in two adults one with a long history of angina pectoris In neither instance was the catheterisation performed with electrocardiographic control Wood (40) records one fatal case of ventricular fibrillation in 233 catheterisations

Conclusions Cardiac catheterisation gives physiological information concerning the heart s action as opposed to angiocardiography which reveals the anatomical condition of the heart and main vessels The determination of cardiac output in health and disease is of value as a physiological experiment and in determining the types of heart failure The chief value now is in the investigation of cases of congenital heart disease in which surgical procedures are contemplated Catheterisation may also help in deciding whether a pulmonary systemic anastomosis or a valvulotomy may give better results in certain types of congenital heart disease A definite clinical diagnosis in any case of congenital heart disease is often very difficult owing to the frequency with which multiple defects are present Holling and Zak (39) state that cardiac catheterisation will enable certain malformations to be proved Thus if the catheter passes through a septal defect it can be seen to do so on the X ray screen On the other hand it must be understood that the passage of a catheter from the right to the left auricle does not prove that there is a functioning defect in the auricular septum In about a quarter of normal subjects the foramen ovale may be patent although functionless allowing the passage of the catheter In these cases the presence of a shunt from one chamber to another can only be proved by blood gas analysis If the catheter tip is passed into the pulmonary artery the pulmonary stenosis can be recognised by the fall in pressure as the tip passes through it An atrial septal defect with an arteriovenous shunt will result in blood in the right atrium being more oxygenated than is the blood in the venæ cavæ Similarly in uncomplicated ventricular septal defect blood samples from the right ventricle especially from the region of the outflow tract will usually show an increase in oxygen content as compared with vena caval and right atrial samples This indicates a left to right shunt between the ventricles With a patent ductus arteriosus the blood in the pulmonary artery is more oxygenated than is the blood in the right ventricle By cardiac catheterisation Eisen

menger's complex may be distinguished from Fallot's tetralogy. The lesions in Fallot's tetralogy consist of stenosis or hypoplasia of the pulmonary artery, a defect in the interventricular septum, the aorta is displaced to the right and communicates with both ventricles, and the right ventricle is hypertrophied. In the Eisenmenger complex there is an increase in the size of the infundibulum, pulmonary valve and artery, together with an interventricular septal defect, enlarged right ventricle and dextroposition of the aorta. In Eisenmenger's complex, therefore, the systolic pressure in the right ventricle equals that in the pulmonary artery, as there is no pulmonary stenosis. Systemic pulmonary anastomosis which is helpful in the Fallot's tetralogy would not be likely to improve the patient suffering from Eisenmenger's complex.

Cardiac catheterisation will also establish the diagnosis of transposition of the great vessels if it is found that the blood in the pulmonary artery is richer in oxygen than the aortic blood. In cases of "pure pulmonary stenosis" or as it is sometimes called "pulmonary stenosis with closed ventricular septum" cardiac catheterisation will decide whether pulmonary stenosis is present. Severe peripheral cyanosis may result from reduced cardiac output without intracardiac shunt. In such a case the arterial blood would have a normal oxygen saturation. Cardiac catheterisation is also used to determine the increase in pressure in the pulmonary artery and right ventricle before the operation of valvulotomy in mitral stenosis. The fall in pressure after operation can be similarly demonstrated.

THIOCYANATES AND THE TREATMENT OF HYPERTENSION

Potassium and sodium thiocyanate were originally recommended for the treatment of essential hypertension by Pauli (43) in 1903. The mode of action is obscure, but it has been suggested that they cause a relaxation of smooth muscle. They are readily absorbed from the gastro-intestinal tract and are distributed throughout body fluids in the same way as chlorides and bromides; they do not penetrate into the cells. They are excreted mainly through the kidneys, but this excretion can vary as much as 400% in different individuals.

Thiocyanates offer one of the few means of treatment of hyper tension but their use was limited owing to toxic effects until the dosage was controlled and rendered more safe by serum estimations Barker and his colleagues (44 45 and 46) who have treated many patients successfully over a number of years emphasise the importance of frequent determination of serum levels of thiocyanates while treatment is in progress They consider a concentration of 8 to 12 mg per 100 ml to be a safe level in the majority of cases They recommend beginning the treatment by giving 0 3 g per day and making blood cyanate determinations every week for four to six weeks and subsequently every month It is readily soluble 0 1 g in ½ oz of water being given three times a day or it may be prescribed as Elixir Sodium Thiocyanate (gr 20 in fl oz 1) one teaspoonful twice a day Enteric sealed tablets containing 0 2 g or 0 065 g of potassium thiocyanate are also sometimes given If the blood pressure drops sharply or if symptoms of fatigue or depression are marked the dose can be decreased If no blood pressure change or toxic symptoms occur after three to four weeks and the blood concentration has not reached 6 mg per 100 ml the dose may be doubled Barker and his colleagues *who reported on 216 cases* found that the systolic pressure may drop during the first two to four weeks whereas the diastolic pressure may not be reduced for three to four month The best clinical results were obtained with patient whose erythrocyte count was high and sedimentation rate low and whose blood pressure was of the fluctuant type Out of 246 patients 76% showed some improvement There were no toxic deaths but patients showed toxic manifestations of one kind or another Another series of 50 cases treated over eleven years was reported by Kurtz Shapiro and Selby Mills (47) These authors followed Barker s method of laboratory control Subjective improvement was very definite in 63% of cases and fair in 17% Satisfactory reduction of blood pressure was obtained in 78% of cases There were no deaths that could be attributed directly or indirectly to thiocyanate Fischmann (48) treated 40 cases with thiocyanate 25 proving successful and 15 being failures Of the successful cases 3 showed no fall in blood pressure but improved symptomatically especially as regards headache Of the 15 failures the blood pressure fell in 6 cases without subjective improvement

To set against these encouraging results there have been some reports of fatalities and toxic effects following the use of thiocyanates. Foulger and Rose (49) in 1943 and Potter (50) in 1944 reported cases of acute goitre due to thiocyanate therapy. Other toxic effects include exfoliative dermatitis, weakness, dizziness, anorexia, thrombo-phlebitis and hæmorrhagic encephalomalacia. The importance of a strict control of dosage based on weekly and later monthly serum determinations and a careful watch for untoward symptoms cannot be over-emphasised. Further, thiocyanate should never be given if there is any evidence of renal failure. In our experience it may not lower the blood pressure or the fall produced is only temporary.

METHONIUM SALTS AND THE TREATMENT OF HYPERTENSION

Drugs of the methonium series have been introduced for the treatment of high blood pressure. Paton and Zaimis (51) pointed out the possible clinical value of hexamethonium iodide (C_6) in hypertension and vascular disease, as it differs from tetraethyl ammonium iodide (T.E.A.) in its greater potency, slower action and more prolonged effect. Observations were next made by *Organe, Paton and Zaimis (52) on the action of pentamethonium* iodide (C_5) on man. On intravenous injection these drugs produce a fall in blood pressure, increase in pulse rate, vasodilatation which *is more marked in the lower than in the upper* limbs, diminution of sweating especially in the feet, dilatation of the pupils with possibly disturbance of accommodation. Nausea, headache and dryness of the mouth may be noted as side effects. The action of the drugs is on the ganglion cells of the autonomic nervous system, both sympathetic and parasympathetic, a blockage of impulses being produced there.

Campbell and Robertson (53) reported on the treatment of eight cases of *hypertension and papilloedema with hexamethonium* bromide. Treatment was commenced with the intramuscular injection of 100 mg every [illegible] hours and subsequently the drug was given *by mouth* by dissolving a 0 [illegible] g tablet in 20 ml of water. The dosage adopted was 0.25 g twice daily on the first day, 0.25 g t.i.d. for the next two days, with a gradual increase up to 0.5 g four to six times a day. The drug is most effective when taken before meals. Good results were obtained, the blood pressure

falling headache disappearing and papilloedema diminishing Certain unpleasant reactions may occur such as blurring of vision dryness of the mouth nausea heartburn and constipation More serious side effects include paralytic ileus distension of the bladder postural hypotension and fainting In addition to the effect in lowering the blood pressure C_6 has a profound influence in diminishing gastric motility and gastric secretion Kay and Smith (54) showed that the intramuscular injection of 100 mg of hexamethonium iodide produced achlorhydria lasting for three hours The action here is by vagal blockade and the drug prevents the development of a true insulin response such as is noted with a complete vagotomy (see p 172) When C_6 is given with insulin the gastric acidity first falls and then rises to little above the resting value C_6 fails to prevent a histamine response of acid gastric secretion for it is thought that histamine acts directly on the parietal gastric mucosal cells Kay and Smith (55) also showed that the iodide and bromide of methonium were more powerful gastric depressants than is the chloride

Pentamethonium iodide (Lytensium) is put up in ampoules for injection containing 100 mg /1 ml Hexamethonium iodide (Hexathide) is also available in ampoules containing 50 mg /2 5 ml The hexamethonium bromide (Vegolysin) tablets contain 250 mg They are soluble in water

In one of our cases of essential hypertension treated by C_6 we found that when hexamethonium iodide was given intravenously in a saline drip the blood pressure fell from 220/130 mm Hg to 140/108 mm Hg after four doses of 10 mg given at intervals of three minutes The pressure began to rise again after one and a half hours Hexamethonium bromide was subsequently given by mouth in doses of 250 mg tablet crushed in water four times a day before meals but the blood pressure was not so well controlled although there were no unpleasant side effects

The dosage should be controlled by blood pressure readings taken weekly as the drug may have a cumulative effect and bromism or iodism may ensue Postural hypotension is one of the most dangerous of the side effects It is especially liable to occur when the patient is standing still Turner (56) records the case of a man who was all right when walking from hospital to the bus stop but the trouble came on when he was standing in the

queue An abdominal belt or binder is a useful corrective and the patient should be instructed to contract his abdominal muscles and if possible lie down if he feels faint An injection of adrenaline should be given in severe hypotensive reactions

Conclusions It is too early to say whether the methonium compounds will prove as disappointing as the various other drugs recommended in the past for the treatment of hypertension They certainly should not be used indiscriminately and without proper control They appear to be of most value in cases of threatened left ventricular failure diminution of vision due to retinal changes impaired renal function and when there appears to be a possibility of a stroke

The absorption of the pentamethonium and hexamethonium salts when given by mouth appears to be very uncertain and in order to obtain an adequate fall in blood pressure as much as 3 to 4 g per day may be necessary Postural hypotension may result from these large doses Further the danger of bromism or iodism is enhanced when the patient is on a low salt diet The drugs are excreted by the kidneys and if there is renal failure much smaller doses must be employed

The Rice Diet in Hypertension

Kempner (57) in 1944 introduced the rice diet for the treatment of patients suffering from renal and hypertensive vascular disease He found improvement in the blood pressure and also in the size of the heart retinal changes and electrocardiograms A clinical trial of the diet was carried out for the Medical Research Council and the report was published in 1950 (58) Patients were encouraged to eat 250 to 350 g of rice (dry weight) daily It was boiled in water or steamed with no added salt milk or fat It can be flavoured with sugar lemon juice or fruit and served dry or wet The fluid was limited to 1 litre in twenty four hours The diet contained sodium [illegible] mg potassium [illegible] mg chloride 195 mg and nitrogen 3 14 g (equivalent to [illegible] g of protein)

A sample menu is as follows

Breakfast Grape fruit (prepared) [illegible] oz boiled rice (dry weight) 2 oz sugar 1 oz and orange juice 8 oz

Mid morning Lemon juice 1¼ oz with water 5 oz and sugar ¼ oz

Lunch Boiled rice (dry weight) [illegible] oz golden syrup ¼ oz blackcurrants (raw) 5 oz sugar ½ oz and black grapes 6 oz

Tea Boiled rice (dry weight) 1 oz sugar 1 oz banana slices 1½ oz orange juice – oz and lemon juice –¼ oz

Supper Boiled rice (dry weight) 3 oz sugar ¾ oz fresh peaches 5 oz and orange juice 8 oz

On retiring Dessert cherries 5 oz

This diet contains C 49– g P –6 g and F 3 g sodium 83 mg chloride 139 mg

The Committee confirmed Kempner s findings of a fall in blood pressure The average fall in 35 patients treated by the Committee over periods ranging from twenty to ninety five days was 55 mm Hg systolic and 26 mm Hg diastolic

It appears probable that the fall in blood pressure is due not to the low protein content but to the low sodium in the diet As soon as the sodium chloride intake is increased even slightly the blood pressure tends to rise The diet is not without danger as uræmia has been recorded during treatment by Schroeder (59) in 1948 If in non œdematous patients the urinary sodium or chloride does not fall to a low level within a few days of starting the diet the Committee emphasise that a careful watch must be kept on the blood urea and blood sodium content

We have tried the diet on patients suffering from heart failure with high blood pressure and œdema and in these cases we have found that the patient is usually too ill to take the diet

Some Cardio therapeutic Measures

The Use of Digitalis Digitalis still remains supreme amongst the drugs used in the treatment of heart disease Withering (60) in 1785 was one of the first to draw attention to the value of digitalis in producing diuresis in certain cases of heart failure Since then the indications for its use have become more clearly defined and it is recognised that its most beneficial effects are obtained in cases of cardiac irregularity due to auricular fibrillation We no longer believe in the *circus* movement but the ventricle is slowed by increasing the degree of heart block It is also of value in auricular flutter and indeed in heart failure from any cause High blood pressure is not now considered a contra indication as it has been shown that in therapeutic doses digitalis does not cause a rise of systolic pressure although the pulse pressure may be increased as the result of a fall in the diastolic level in cases

of cardiac failure Whether or not it acts as a direct *cardiac tonic* and should be employed in disorders associated with sinus rhythm has been a matter of dispute It is usually held that digitalis in such cases is a cardiac tonic and slows the whole heart by its action on the vagus thus prolonging diastole increasing the force of systole and improving the coronary circulation and so the nutrition of the cardiac muscle

In heart failure of a severe degree large doses are usually required On account of the danger of producing severe toxic effects the total quantity necessary is generally administered in divided doses at several hours interval but the beneficial effect is thereby inevitably postponed This delay is partially due to the fact that digitalis takes about six hours before it is absorbed from the stomach it is also eliminated slowly by the intestines and kidneys and so cumulative results may occur

Digitalis may be administered either in the form of a good tincture or as a tablet of the dried leaves One cat unit tablet corresponds approximately with 1 ml (m 17) of the tincture or 0 1 g (gr 1 5) of tab digitalis pulv (B P Add) In order to obtain results as quickly as possible in cases of severe heart failure the maximum amount of digitalis which can safely be administered to an individual should be given within twenty four hours This may be taken to be m 17 of the tincture or gr 1 5 of the dried leaf for every 10 lbs of body weight Thus for a man weighing 10 stones the amount required is m 240 of the tincture or gr 21 of the dried leaf tablet When the weight of the patient is not known accurately as *will often be the case* three quarters *of the* above amounts should be the maximum given Digitalis should not be administered more often than every six hours Thus in an urgent case in which no digitalis has been taken within the last ten days m 60 of the tincture may be given six hourly for 4 doses and then the dose reduced to m 30 if digitalisation has not yet been effected As soon as this occurs the drug should be discontinued for a day or so and then m 20 of the tincture or less may be required three times a day to allow for daily elimination and to keep the patient under the influence of the drug If digitalis has been administered within the last ten days the total calculated amount should be *reduced to three quarters if no signs of digitalis* over action are shown by the electrocardiogram such as heart

block or premature ventricular systoles and to one half if they are present In either case the total calculated amount is given in three equal doses at six hourly intervals

In conclusion it may be said that massive doses should only be used in cases of heart failure of extreme urgency and generally administration in smaller doses is the safest course to adopt

The indications for discontinuing the drug are according to Eggleston Nausea or vomiting unless this is due to venous congestion of the stomach which is usually the case when it occurs shortly after the commencement of digitalis administration (Jensen (61)) The apex rate lower than 60 The occurrence of frequent ectopic systoles heart block phasic arrhythmia or coupled beats The latter are caused by a regular sequence of premature ventricular systoles following each normal ventricular contraction

It is usually found if the digitalis is going to relieve the heart failure that diuresis takes place if there is no diuresis beneficial results are not likely to ensue and diminution in the secretion of the urine is an additional sign of digitalis toxæmia The diuresis is a manifestation of the effect of digitalis in filling the arteries and emptying the veins and is due to an increased renal circulation and relief of renal congestion Diuresis does not follow the administration of digitalis even in massive doses unless œdema is present

Digitalis should not be administered more frequently than at six hourly intervals as this is the time taken for its absorption from the stomach and in cases of auricular fibrillation it has been shown by Canby Robinson (62) that an effect is produced in two to five hours after oral administration Sufficient warning of any toxic effects which may be produced by its administration is thereby given

Digoxin Wokes (63) in 1929 showed that the leaves of the foxglove *Digitalis lanata* are physiologically nearly four times as potent as are those of *Digitalis purpurea* the species used in medicine Smith (64) in 1930 isolated a pure crystalline alcohol soluble glucoside Digoxin from the leaves of the *Digitalis lanata* It possesses the advantage that it does not require biological standardisation It is absorbed and excreted more quickly than is digitalis and can be given orally or intravenously Intravenous injection is only advisable when an extremely rapid effect is

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obtained in fibrillation of less than six months duration and in those cases in which the symptoms increase with the onset of fibrillation The infective group of cases is more suitable than the degenerative The drug can be tried in paroxysmal auricular fibrillation as digitalis does not usually have any beneficial effect here (Parkinson and Nicholl (69)) It may be of value in auricular flutter the normal rhythm being restored direct without an intermediate stage of fibrillation It is at times successful in those cases of flutter which have been converted to fibrillation by the use of digitalis but which do not return to the normal sinus rhythm on discontinuing the drug It does not appear to have any beneficial effect on premature systoles

Contra indications There are certain definite contra indications which apply no matter what the nature of the cardiac disorder may be Thus quinidine must not be given if the heart is greatly enlarged When there is congestive failure the patient should first be treated with rest digitalis and diuretics until compensation is restored Then after a week s cessation from the use of digitalis quinidine may be given if thought advisable Heart block is also a contra indication It must not be given if there is a history of recent embolism It is considered inadvisable to administer quinidine if fibrillation has lasted for six months or to use quinidine and digitalis simultaneously

Regimen The patient must be kept in bed and preferably at absolute rest during the initial week of the treatment This usually implies the necessity for skilled nurses Attention must be paid to the bowels and the diet should be such as will not give rise to flatulence

Methods of Administration and Dosage The drug is given by mouth in powder form in gelatine capsules Probably the safest method is to give on the first day a test dose of 0 2 g (3 gr) to determine whether the patient has any idiosyncrasy to the drug If no toxæmic symptoms appear on the second day 0 4 g (6 gr) is given every three hours for four doses and this is increased to five doses of 0 4 g at three hourly intervals on the third day This dosage is then maintained until the end of the week and if the normal rhythm has not been restored by this time the drug will probably not prove successful The pulse should be taken before each dose is given and if it is found to be regular the drug

required or when the patient vomits anything taken by mouth It must never be given intravenously if the patient has taken digitalis during the previous two weeks In auricular fibrillation the average initial intravenous dose for an adult of 10 stones or over is 0 75 to 1 mg A solution is obtainable in ampoules each ampoule contains 0 5 mg Digoxin in 1 ml 70% alcohol It must be diluted before use with 9 times the volume of freshly prepared sterile normal saline solution The solution thus obtained is injected slowly into a vein taking three to five minutes over the injection The solution is very irritant and must not be injected outside the lumen of the vein Wayne (65) has shown that the ventricular rate rapidly slows after such an injection and that the maximum effect is obtained in one to two hours Subsequently Digoxin is administered by mouth as described below

For oral administration the initial dose is 1 5 mg for patients weighing 10 stones or more and 1 to 1 25 mg for lighter patients Digoxin must never be given by mouth if the patient has taken digitalis during the previous week It is put up in Tabloid form each containing 0 25 mg to be taken in water or in 30 ml bottles containing 0 5 mg in each millilitre of 80 / alcohol The solution is best given in chloroform water to disguise the bitter taste After oral administration the ventricular rate begins to slow in about an hour and the maximum effect is obtained in about six hours Six hours after the initial dose 0 25 mg is given every six hours until the ventricular rate falls to 60 or 70 Subsequently a maintenance dose of 0 25 mg twice a day may be required indefinitely

Quinidine This is a dextro rotatory cinchona alkaloid which was first used by Frey (66) in 1918 in cases of auricular fibrillation. Wenckebach (67) had previously in 1914 obtained a temporary arrest of auricular fibrillation in one patient by giving 1 g of quinine daily Frey in a series of 22 cases of fibrillation obtained a restoration of normal rhythm in 50 /o He gave the drug in doses of 0 4 g (6 gr) t d s by mouth for three to eight days

The administration of quinidine sulphate may be considered under the following headings —

Selection of Case Quinidine is of greatest value in cases of auricular fibrillation especially when there is neither cardiac enlargement nor valvular disease (Cotton (68)) Best results are

The Action of the Drug The effect of quinidine upon heart muscle can be summarised thus The auricular rate is slowed The rate of conduction in the auricle ventricle and A-V junctional tissues is reduced The absolute refractory period of auricular muscle is prolonged

Quinidine has also some effect upon the vagus nerve causing a partial paralysis which tends to increase the ventricular rate

The electrocardiographic changes which may occur during quinidine administration are according to Hay (70) Slowing of auricular rate the onset of auricular flutter and 2 1 heart block increase of ventricular rate appearance of ectopic ventricular beats restoration of normal rhythm in 50 / of cases of fibrilla tion

The Value of the Drug The administration of quinidine has very definite dangers and the cases in which it is worth a trial require careful selection and very close watching during the treatment Even if normal rhythm is established the general condition of the patient is not always improved while the under lying myocardial degeneration still remains

Mercurial Diuretics and Ammonium Chloride These drugs include mersalyl Neptal Mercupurin Mercuhydrin Novurit and Esidrone They are of great value in the treatment of congestive failure (right ventricular failure) and in acute œdema of the lungs (left ventricular failure) Mersalyl is an organic mercurial compound which was reported on clinically by Brunn (73) and by Bernheim (74) in 1924 It contains 36 / of mercury and is put up in 10 / solution in 1 ml and 2 ml ampoules Mersalyl has now replaced novasurol which was introduced by Saxl and Heilig (75) in 1920 Novasurol has been abandoned owing to its toxic effects whereas mersalyl is not usually toxic Andrews (76) reported a case of heart failure without renal disease in which the first intravenous injection of 0 5 ml of mersalyl re ulted in headache epileptiform convulsions and unconsciousness but the patient a woman aged twenty two did not die In the case described by Higgins (77) there was hypertensive heart disease with congestive failure About 10 doses of mersalyl or Mercupurin had been given when an intra muscular injection of 1 ml of Mercupurin was followed an hour later by a severe chill fever dyspnœa cyanosis fall in blood pressure and prostration Death seemed imminent A month

should be discontinued at any rate temporarily until a tracing has been taken Further indications for discontinuing the administration of quinidine are a slowing of the auricular rate to about 250 per minute or an increase of the ventricular rate to over 160 In the former case there is a danger of the ventricle following directly the auricular rate The exact rate of the auricle and ventricle are best determined by an electrocardiogram

Hay (70) recommends that quinidine should be given every two hours by day in ten equal doses as its action soon passes off Starting with a test dose of 0 2 g (3 gr) on the first day 2 g (30 gr) are given on the second day in ten doses of 0 2 g each If necessary the dose may be increased to 3 g (45 gr) a day in ten doses of 0 3 g (4 5 gr) The smallest beneficial dose appears to be one of 5 gr which restored the normal rhythm in a case of auricular fibrillation of recent origin under our care which was associated with a toxic goitre As much as a total of 375 gr has however been required before a satisfactory result was obtained and it may be necessary to continue indefinitely with small doses of 5 gr daily especially if a relapse occurs on omitting the drug

Symptoms of Toxæmia and Dangers Frey (66) in 1918 drew attention to the possibility of quinidine idiosyncrasy resulting in such symptoms as respiratory failure and cerebral paralysis Benjamin and V Kapff (71) in 1921 recorded instances of embolus *occurring during the administration of quinidine This is especially* liable to follow restoration of normal rhythm and is in all probability due to the detachment of a clot from the wall of the auricle Embolus formation is not necessarily fatal The onset of ventricular fibrillation may cause sudden death

Headache is nearly always experienced while quinidine is being taken Other toxic symptoms include sweating nausea vomiting diarrhoea abdominal pain and a scarlatiniform rash Dimness of vision or the occurrence of frequent ectopic ventricular beats is an indication for the immediate discontinuance of the drug

Results of Quinidine Administration Campbell and Gordon (72) found *that quinidine restored normal rhythm in 64% of a series* of 135 cases of auricular fibrillation In 30 of these however fibrillation recurred after an average period of two years but in the other 34 % the normal rhythm was still maintained after an average period of nearly four years

cases we have found that the administration of ammonium chloride alone will produce a good diuretic response.

It is important that the weekly injections of mersalyl with the corresponding administration of ammonium chloride should be continued for an indefinite period providing a good diuresis ensues and that the patient does not become unduly dehydrated. We have patients who have received this treatment for two years or longer and who are still continuing with it thereby being enabled to maintain a fair degree of health and activity. In other cases we have noticed that dehydration appears excessive after a prolonged course—the patient feeling weak and ill after each injection. Here the injections should be discontinued and if required again later they should be given at longer intervals. Rechtschaffen and Gittler (80) record a case of acute dehydration following the subcutaneous administration of Meralluride (Mercuhydrin) Sodium. Within four hours of the initial dose the patient suffered from peripheral vascular collapse bradycardia elevation in temperature and a shaking chill. Recovery occurred within two hours. During the four hours after the injection there was a diuresis of 1 800 ml. It should be remembered that as much as 8 to 10 pints of excess fluid may be retained in the body tissues without pitting œdema being apparent. Such a condition may be termed latent œdema and is associated with pulmonary congestion and œdema of the lungs and with an engorged liver. It is much relieved by the administration of mersalyl or Novurit.

The Circulation Time

Various methods have been employed to estimate the circulation time in man. Bornstein (81) in 1912 made the patient breathe a mixture of air containing 5 to 7% CO_2. The time taken for the arterial blood to carry an increased amount of CO_2 to the respiratory centre was shown by an increased depth in the breathing. Since then various substances have been injected intravenously such as fluorescein histamine calcium chloride saccharin ether sodium dehydrocholate calcium gluconate and magnesium sulphate. Tarr Oppenheimer and Sager (82) in 1933 gave an historical account of the methods which have been used and reported their experiences with sodium dehydrocholate (Decholin Sodium). This measures

later the patient died and the kidneys showed some mild arteriolo sclerosis with very slight necrosis and calcification of the tubules probably due to mercury Volini et al (78) reported 3 cases of very sudden death following the intravenous injection of mercurial diuretics 1 from *Mercupurin* and 2 from *Esidrone* In 2 of the cases electrocardiograms showed the development of ventricular fibrillation Evans and Perry (79) have also published 6 cases of immediate death due to the same cause and suggest that it may be advisable to give mercurial diuretics intramuscularly to patients with a low plasma protein In other cases cramps may develop in the legs due to depletion of chlorides and these can be relieved by the administration of sodium chloride Mercurial diuretics should not be given in cases of acute nephritis in lipoid nephrosis or if the urine contains some granular or blood casts or if there is hæmaturia A test dose of 0 5 ml mersalyl is first injected intra muscularly and if there is no intolerance as shown by hæmaturia diarrhœa or irritation of the skin a course of intravenous injections is begun In order to render the urine acid ammonium chloride in doses of gr 30 is given by mouth every six hours The ammonium chloride can be administered as a mixture Ammon. chlor gr 30 ext glycyrrhiz liq m 20 sp chlorof m 7 aquam ad 1 oz or in the form of four 0 5 gram stearettes On the third day of the administration of the ammonium chloride 1 ml of mersalyl diluted with 4 ml of sterile normal saline is injected slowly into a vein in the arm The arm is then raised to prevent venous thrombosis The ammonium chloride is administered daily On the fifth and ninth day 2 ml of mersalyl diluted with 8 ml of sterile normal saline are injected intravenously If a good diuretic response is obtained such as 90 to 180 oz of urine the injections of mersalyl should be repeated at intervals of a week the ammonium chloride being given by mouth for two days before the day of and the day after the injection i e for four days each week The total fluid intake during the twenty four hours must be restricted to 35 oz or less and full digitalisation should be maintained to control the rate of the heart The fluid intake and output should be recorded each twenty four hours and the patient weighed at first daily and later weekly The degree of dehydration is reflected in the loss of weight No salt should be used in the cooking of the food and none added when it is eaten In some

serial readings are of value as the circulation time in an individual varies according to changes in his condition being prolonged when he is worse and vice versa They also found it of value in determining whether there is or is not cardiac failure in the presence of emphysema *i e* whether the dyspnœa is predominantly pulmonary or cardiac If the circulation rate is rapid then the heart is well compensated In 7 cases of bronchial asthma the arm to tongue circulation time was found to be shortened The ether circulation (arm to lung time) is measured by injecting into an antecubital vein 5 m of ether well shaken with 5 m of normal saline using a tuberculin syringe and a large needle The time is recorded from the moment of injection to the moment when the patient or the observer smells the ether in the breath The average normal time is 5 7 seconds (extremes ? 6 to 10 seconds) The arm to tongue time measures roughly the circulation rate as a whole the arm to lung time indicates the functional capacity of the right ventricle In right sided failure therefore the ether time is prolonged the lung to tongue time (the Decholin or the magnesium sulphate time minus the ether time) is a test for the functional capacity of the left ventricle Thus Hitzig (84) showed that in isolated failure of the left ventricle the arm to tongue circulation time is almost always prolonged sometimes up to three times its normal value but the arm to lung time may be simultaneously normal Occasionally however with severe left sided failure the arm to tongue circulation time is normal possibly owing to a very powerfully acting right ventricle Cottrell and Cuddie (85) have used the Decoholin method to investigate the circulation time in 1 cases of bronchial asthma and found it to be normal both during and between the attacks

Conclusion In our experience using the Decholin method the arm to tongue circulation time has proved of value in distinguishing between cardiac and pulmonary dyspnœa especially in cases of emphysema or bronchial asthma complicated by auricular fibrillation myocardial degeneration or a valvular lesion

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the arm to tongue *circulation time* The test is carried out before breakfast with the patient lying as flat as possible the right arm being supported at the level of the auricles Five ml of a 20% solution of Decholin are injected through a wide bore needle into an antecubital vein taking one to two seconds over the injection The patient is told to raise his left hand directly he perceives a bitter taste under the tongue *The time is taken with a stop watch from the beginning of the injection of the Decholin to the signal* that it has reached the tongue The average normal time is thirteen seconds (extremes ten to sixteen seconds) Occassionally the patient complains of nausea after the injection or of pain in the region of the gall bladder The circulation time is not affected by the weight height pulse rate or blood pressure of the patient *It is prolonged in congestive heart failure in the majority of cases to between* twenty and forty seven seconds The circulation time is also prolonged in myxœdema and polycythæmia but shortened in fevers anæmia and hyperthyroidism when the latter is not complicated by heart failure Bernstein and Simkins (63) in 1939 used magnesium sulphate to determine the arm to tongue circulation time *Six ml of a 10% solution of magnesium* sulphate are *injected* intravenously into an antecubital vein and when it reaches the mouth the patient experiences a transient hot sensation in the pharynx and tongue which spreads rapidly to the face hands perineum and legs It is advisable to have at hand an ampoule of 10 calcium gluconate for immediate intravenous injection should an undue magnesium sulphate reaction occur *With this method Bernstein and Simkins found the average arm to tongue* circulation time *12 9 seconds (extremes 7 to 1" 8 seconds) They classified cases of heart failure clinically into groups* (1) well compensated (2) with dyspnœa on exertion (3) with dyspnœa and cyanosis (4) with additional pulmonary congestion (5) with ascites or œdema In the majority of cases the circulation time was prolonged proportionately to the degree of heart failure up to *about thirty* seconds Lesions such as *mitral* stenosis or *auricular fibrillation if not accompanied by decompensation* did not affect the circulation time *The authors state however* that the circulation time is not the *infallible* diagnostic tool it was once supposed to b and it is not really uncommon to find a normal circulation time in a markedly decompensated patient However

serial readings are of \alue as the circulation time in an indi\idual \aries according to changes in his condition being prolonged when he is worse and \ice \ersa They also found it of value in deter mining whether there is or is not cardiac failure in the presence of emphysema i e whether the dyspnœa is predominantly pulmonary or cardiac If the circulation rate is rapid then the heart is well compensated In 7 cases of bronchial asthma the arm to tongue circulation time was found to be shortened The ether circulation (arm to lung time) is measured b\ injecting into an antecubital \ein 5 m of ether well shaken with 5 m of normal saline using a tuberculin syringe and a large needle The time is recorded from the moment of injection to the moment when the patient or the obser\er smells the ether in the breath The average normal time is 5 7 seconds (extremes 2 6 to 10 seconds) The arm to tongue time measures roughly the circulation rate as a whole the arm to lung time indicates the functional capacit\ of the right \entricle In right sided failure therefore the ether time is prolonged the lung to-tongue time (the Decholin or the magnesium sulphate time minus the ether time) is a test for the functional capacity of the left \entricle Thus Hitzig (84) showed that in isolated failure of the left \entricle the arm to tongue circulation time is almost always prolonged sometimes up to three times its normal \alue but the arm to lung time may be simul taneousl\ normal Occasionally howe\er with se\ere left sided failure the arm to tongue circulation time is normal possibl\ owing to a \ery powerfully acting right \entricle Cottrell and Cuddie (85) ha\e used the Decoholin method to in\estigate the circulation time in 1 cases of bronchial asthma and found it to be normal both during and between the attacks

Conclusion In our experience using the Decholin method the arm to-tongue circulation time has pro\ed of \alue in distin guishing between cardiac and pulmonary dyspnœa especially in cases of emphysema or bronchial asthma complicated by auricular fibrillation myocardial degeneration or a valvular lesion

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Longcope (2) observed areas of deep red moist solidification in the lungs with purulent material in the bronchi The alveoli in the affected areas were filled with coagulated serum containing red corpuscles and mononuclear cells polymorphonuclears being rarely seen Campbell et al (3) consider that the primary process is a peribronchitis peribronchiolitis bronchitis and bronchiolitis with swelling of the bronchial epithelium and exudation into the bronchi and bronchioles resulting in œdema absorption of the contained air and localised areas of collapse The alveolar walls become thickened and the alveoli contain mononuclears Areas of encephalitis were also seen Perrone and Wright (4) reported a fatal case of primary atypical pneumonia complicated by encephalitis the patient dying on the fourteenth day of the illness Foci of increased cellularity were seen in the cerebrum cerebellum basal ganglia and paraventricular tissue The cells forming these foci were astrocytes and microcytes There were also areas of lymphocytic perivascular infiltration in the meninges

Clinical and Laboratory Findings The incubation period varies from two to twenty one days or longer The onset is frequently insidious with malaise severe headache a racking dry cough and muscular pains in the limbs and abdomen Anorexia may be a marked feature and at times the patient complains of pain in the chest and dyspnœa or the illness may begin with an upper respiratory tract catarrh Some cases are apyrexial the patient having a persistent cough with catarrhal symptoms and the nature of the disease is only recognised when the lungs are examined radiographically The temperature usually rises to 100 or 103 F and after remaining at about this level for eight to ten days falls by lysis although secondary rises of temperature are sometimes met with

The sputum is usually mucoid and at times contains a few streaks of blood or small hæmoptyses of a teaspoonful or so occur but the expectoration is never rusty or of a prune juice appearance It contains no predominating or characteristic organisms but mononuclear cells may be seen The physical signs in the lungs are indefinite and variable small areas of collapse indicated by slight dullness and weak air entry may appear from day to day The most characteristic finding is the presence of showers of sticky rales heard especially after cough over the

CHAPTER IX

THE LUNGS

Primary Atypical Pneumonia

(Virus Pneumonia)

Owing perhaps to the greater facilities for, and importance attached to radiographic examination of the lungs attention has been increasingly directed during recent years to certain catarrhal pulmonary conditions usually of a benign character in which the X ray films show soft shadows in the lung fields The term primary atypical pneumonia has been used to cover a group of allied diseases such as virus pneumonia pneumonitis influenzal pneumonia broncho pneumonia of unusual or undetermined aetiology acute diffuse bronchiolitis acute bronchiolitis with atelectasis, and atypical pneumonia with leucopenia When we have excluded conditions of which the cause is thought to be known such as influenza due to Pfeiffer s bacillus or to the viruses A B or X associated with staphylococci or streptococci the pneumonic consolidation of psittacosis Rickettsia infections such as the Q fever of Australia American Q fever and ornithosis we are left with a group of cases in which the cause is unknown but is probably a virus and which may be designated as primary atypical pneumonia virus pneumonia or virus pneumonitis

Primary atypical pneumonia tends to appear in small epidemics and probably also exists endemically It is not highly contagious but may infect doctors and nurses looking after cases in hospital wards Young adults are chiefly affected and there is no particular seasonal incidence The disease has not been transmitted to animals but Bragg (1) in 1945 successfully conveyed it to human volunteers by spraying the nose and throat with a bacterium free filtrate obtained from the throat washings and sputum collected from patients suffering from atypical pneumonia Thus 3 out of 12 men receiving this inoculum developed the disease twelve to fourteen days later and cold agglutinins for group O human red cells were found in their serum in titres varying from 1/128 to 1/1 024 twenty to twenty seven days after infection

The mortality rate of the disease is low about 0 2% and few observations have been published on the post mortem findings

site for the shadow and owing to areas of atelectasis the dia phragm may be raised on one side and the mediastinum corre spondingly displaced Complications are rare the violent cough has been thought to give rise to multiple fracture of ribs and encephalitis is a dread development Venous thrombosis in the legs and polyarthritis have been occasionally described

Cases may be mistaken for pulmonary tuberculosis especially when the X ray shadows are situated near the apex and in other instances an erroneous diagnosis of primary atypical pneumonia has been made on radiographic evidence when the patient was in reality suffering from lung abscess or bronchial carcinoma At times the prolonged continuous type of fever with a relatively slow pulse rate and low white cells count suggests enteric fever In influenza no abnormal shadows are likely to be seen on X ray examination of the lungs Difficulty may also arise with cases of pneumonia which have been treated with sulphonamides when the signs in the lungs persist there is no leucocytosis and the X ray shadow has a reticulated appearance Further an area of basal bronchiectasis with surrounding inflammatory changes may lead to diagnostic difficulty The lesions of primary atypical pneumonia do not give rise to subsequent pulmonary fibrosis

Treatment It is most important to keep the patient in bed until the temperature has been normal for a week and he should not resume work until the sedimentation rate of the red cells is normal and the X ray shadows have disappeared Penicillin streptomycin and the sulphonamides are useless and the latter may be dangerous giving rise to a hæmolytic crisis Dramatic results are obtained with the use of aureomycin We have seen the tem perature fall to normal within thirty six hours of commencing treatment with a rapid improvement in the patient s condition The dosage recommended is given below in the article on Q fever Steam inhalations may relieve the distressing cough but it is wise to avoid the use of opiates except in small doses For severe cyanosis and dyspnœa oxygen should be administered by nasal catheter or the B L B mask

Q FEVER IN GREAT BRITAIN

Derrick (8) in 1937 first described Q fever in Queensland Australia and by injecting infected blood and urine into guinea

affected portions of the lungs In some instances there is dry pleurisy and rarely a pleural effusion develops The pulse rate is often lower than would be expected from the temperature In more severe cases there is marked dyspnoea and cyanosis the temperature is high and irregular the patient being severely prostrated The white cell count is usually normal but there may be a slight leucopenia The presence of a leucocytosis would indicate the advisability of reconsidering the diagnosis The blood Wassermann reaction *may be positive for a short period* and *during the acute stage of* the illness the sedimentation rate of the red cells is increased to between 20 and 40 mm plasma at one hour

Peterson Ham and Finland (5) described the presence of cold agglutinins in high titre in the serum of patients suffering from primary atypical pneumonia and this observation has been confirmed by Horstmann and Tatlock (6) and others In the majority of cases the reaction is positive between the second and fourth weeks *of the illness Dilutions of the patient s serum are made* from 1/4 to 1/4 096 in 1 ml volume and to each tube is added 0 1 ml of a 2 % suspension of washed group O human erythrocytes The tubes are kept at 0 C overnight and readings made immediately on removing them from the ice box and again after they have stood at room temperature for several hours to see whether the reaction is reversible at room temperature and so indicative of the presence of a true cold agglutinin The test is not entirely *specific as it has been observed in African trypanosomiasis*

Although there are no typical X ray findings the diagnosis cannot be made in the absence of abnormal shadows in the skiagram of the lungs *There is often a marked discrepancy between* the extent of the clinical and radiological findings the latter being more extensive further the shadows in the film frequently persist for some time after convalescence is established and the patient is afebrile Patchy opacities may be seen somewhat resembling those cast by areas of bronchopneumonia They are often irregular in shape and vague in outline and may have a hazy or ground glass appearance migrating in position from day to day At times *the affected areas present a reticulated appearance* Campbell et al (3) found that 81 of the lesions were situated at the base each side being equally affected Drew et al (7) observed that the costo-phrenic angle *especially on the left side is a common*

with lobar consolidation pleural friction or pleural exudation The spleen may be palpable Derrick described conjunctival congestion in 2 of his original 9 cases X ray examination of the lungs shows patchy rather uniform shadows which may persist after the patient is apparently recovered Rickettsial affections in man are usually accompanied by skin rashes but this is not so in Q fever There is no naso pharyngeal catarrh The temperature often falls by lysis after a week but the febrile period may be prolonged to three or four weeks There is usually no anæmia and the white cell count is generally within normal limits although there may be a relative or absolute lymphocytosis The virus was found in the blood of 8 of Derrick s cases but in only 3 cases in the urine The serum taken by Derrick on the twelfth and twentieth days of the disease showed an increase in agglutination titre with a rickettsial emulsion from 1 in 4 to 1 in 20 The death rate is low some authorities placing it at 1 in 500 cases and the prognosis is worse in patients after the age of fifty

MacCallum and his co workers (12) who investigated the cases described by Harman concluded that the nurse two pathologists and the post mortem attendant who were victims of the disease were all infected from the same patient The incubation period in the case of the post mortem attendant was fifteen days which agrees with Derrick s figure of fifteen days or less They further found an endemic focus of Q fever at a town in the South East of England *R burneti* being isolated from the raw milk from some of the farms in the neighbouring area although the cows appeared to be healthy Whittick (13) reported on the post mortem findings in the fatal case in Harman s series Rickettsiæ were found in the lungs spleen testis brain and kidneys The micro scopical appearances of the lungs resembled those found in psittacosis and virus pneumonia

Manderson (14) described a case in Bristol in 1949 which pre sented the clinical features of Q fever and in which specimens of serum taken on the twenty sixth and thirty ninth days of illness gave a positive agglutination in a titre of 1 in 80 This was con sidered to be diagnostic and as the first specimen was obtained over three weeks after the onset of the illness a rise in titre was not to be expected Further observations on the strains of *R burneti* isolated in Great Britain have been made by Marmion and Stoker (15)

pigs Burnet and Freeman (9) found the causative agent to be a rickettsia the *Rickettsia burneti* Derrick described 9 cases occurring among employees in a large meat works and said the name Q fever was chosen until a fuller knowledge should allow a better name Q fever has subsequently been recognised in Europe Africa and America Specific agglutination or complement fixation reactions are given by the rickettsia with serum from Q fever convalescents the antibodies persisting for several years The *R. burneti* has been found in the bandicoot a small bush animal and in various ticks It was first thought that infection occurred in man by inhaling dust containing dried tick excreta It was subsequently shown that occupational exposure to dairy cattle and drinking raw milk from infected cows were the most likely sources of human infection The cows do not appear ill but have a chronic infection of the udder from which the milk becomes contaminated

Stoker (10) in 1949 working in Cambridge tested specimens of serum from patients diagnosed as suffering from atypical pneumonia and found Q fever complement fixing antibodies in 3 specimens out of 24 examined Two of these patients had never been overseas and this was regarded as evidence that Q fever occurs in the British Isles Later in 1949 Harman (11) described 8 cases of Q fever treated at the Royal Cancer Hospital London The diagnosis was confirmed by serological tests and by isolating the *R burneti* in one case

Clinical and Laboratory Findings The disease is characterised by a sudden onset The temperature often rises rapidly to 103 or 104 F It may fall to normal once or twice in the twenty four hours and this is thought not to be due entirely to taking anti pyretics The pulse is slow and there are often heavy sweats with rigors in severe cases Prostration delirium and even coma have been described Headache both retro orbital and occipital is severe together with generalised muscular aching Within a few days a dry irritating cough appears which aggravates the headache Occasionally a little blood stained sputum is expectorated Chest signs although slight can be found on careful examination there are small areas in the lungs of high pitched bronchial breathing with fine rales suggestive of localised alveolar exudation with lobular collapse or consolidation A few cases have been described

Norway Finland Guernsey and America the former method has been adopted The cattle are tested by the injection of tuberculin and the positive reactors including those infected with tuberculosis are disposed of either by slaughtering or by removal from the herd The animals are retested with tuberculin every six months In this way attested herds are formed which are free from tuberculosis and yield a pure milk supply Area eradication is the most effective method of stamping out the disease in cattle All the herds in regions large and small must be attested In this country a certificate of attestation is issued by the Ministry of Agriculture and Fisheries and according to the quarterly statement of the Ministry of March 1950 the numbers of attested herds were England 20 568 Wales 14 204 and Scotland 12 527 It is probable that in Great Britain about 20 % of milk comes from attested herds whereas the average incidence of tuberculosis in dairy cows not so attested is 20 to 25 %

The designations in force in connection with the sale of milk in this country are as follows —*Tuberculin tested milk* This is milk from cows which pass a periodical veterinary examination and tuberculin test If the milk is bottled on the farm it may be described as *Tuberculin tested milk* (certified) This milk may also be pasteurised *Accredited milk* This is raw milk from cows which pass a periodical veterinary examination but not a tuberculin test It must satisfy the same bacteriological tests for cleanliness as raw tuberculin tested milk *Pasteurised milk* This is milk which has been retained at a temperature of 145 to 150 F for at least thirty minutes or which has been retained at a temperature of not less than 16? F for at least fifteen seconds It must pass a methylene blue reduction test for cleanliness and keeping quality and a phosphatase test to indicate whether the heat treatment has been adequately carried out

In order to stamp out in addition to tuberculosis brucella and other infections conveyed by milk the only safe way is that of universal pasteurisation to which unfortunately there is a good deal of opposition at the present

Infection by Human Tubercle Bacilli

It used to be taught that nurses and doctors and others in contact with open cases of pulmonary tuberculosis were not in danger of

Differential Diagnosis Derrick considered the possibility of the following conditions when he was investigating his cases — Typhus fever undulant fever typhoid and paratyphoid fevers, leptospirosis and influenza It is probable that cases in this country have been mistaken for virus pneumonia

Treatment Apart from symptomatic treatment for relief of headache the drug of choice is aureomycin It should be given in capsules by mouth each capsule containing 250 mg Smaller capsules of 50 mg are made which can be used for children The daily dose recommended is 15 to 20 mg per kg body weight For an adult 250 mg is given every six hours for the first twenty four hours then 500 mg every six hours for two days and then 1,000 mg every six hours until the temperature has been normal for forty eight hours The response to aureomycin is often dramatic the temperature rapidly falling to normal the headache vanishing the sweating ceasing and the patient feeling well and asking to get up In very severe cases the temperature may not fall to normal for over ten days Toxic reactions are rare but nausea and vomiting may occur which can usually be relieved by alkalis

The Prevention of Tuberculosis

The large number of cases of tuberculosis in this country and abroad constitutes a challenge to the public health authorities and invites us to consider what steps can be taken to control and eradicate the disease There are two types of tubercle bacillus which affect man the bovine and the human Bovine tuberculosis is a disease of cattle and children are its chief victims the source of the infection being milk or butter containing living bovine bacilli

Bovine Tuberculosis

Childhood tuberculosis due to bovine infection involves especially the glands bones and intestines although sometimes the lungs are affected both in children and in adults Bovine tuberculosis could be completely eradicated by the application of efficient public health measures to ensure a pure milk supply and in certain countries such as America and Scandinavia this has been very largely effected This desirable result can be achieved in two ways either by stamping out the disease from the dairy herds or by killing the tubercle bacilli in infected milk by pasteurisation In

Norway, Finland, Guernsey and America the former method has been adopted. The cattle are tested by the injection of tuberculin and the positive reactors, including those infected with tuberculosis, are disposed of either by slaughtering or by removal from the herd. The animals are retested with tuberculin every six months. In this way "attested herds" are formed which are free from tuberculosis and yield a pure milk supply. Area eradication is the most effective method of stamping out the disease in cattle. All the herds in regions large and small must be "attested". In this country a certificate of attestation is issued by the Ministry of Agriculture and Fisheries, and according to the quarterly statement of the Ministry of March 1950 the numbers of "attested" herds were England 20,568, Wales 14,204 and Scotland 12,5 7. It is probable that in Great Britain about 20% of milk comes from "attested" herds, whereas the average incidence of tuberculosis in dairy cows not so attested is 20 to 25%.

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Infection by Human Tubercle Bacilli

It used to be taught that nurses and doctors and others in contact with open cases of pulmonary tuberculosis were not in danger of

contracting the disease and that adult pulmonary tuberculosis was in the large majority of cases due to a reactivation of a child hood infection Such a view has fortunately been discredited as it engendered a false sense of security and led to a neglect of adequate precautions It is now universally recognised that pulmonary tuberculosis is a herd disease transmitted usually by the inhalation of tubercle bacilli Those closely associated with tuber culous patients may become infected from the patients cough by droplet infection or by contamination of the fingers by touching objects infected with drying sputum Nurses when making beds may inhale dust containing tubercle bacilli disturbed and scattered in the air when the blankets and sheets are shaken Children in homes where one or both parents suffer from open tuberculosis are almost certain to become infected The same applies to children taught by a schoolmaster or schoolmistress who has open ' disease or who are looked after by a nurse similarly affected By an ' open case is meant one in which tubercle bacilli are found in the sputum

The Mantoux Test

This test enables us to determine when children and young adults become infected with tuberculosis It is performed by injecting into the skin of the forearm 0 1 ml of old tuberculin (O T) diluted to 1/10 000 The arm is observed forty eight hours later and a positive reaction consists of a central zone of oedema not less than 5 mm in diameter surrounded by a zone of erythema If the test is negative it is repeated twenty four hours later using now 0 1 ml of 1/1 000 O T If this is negative a third test is made with 0 1 ml of 1/100 O T *The first test should be made in the lower part of* the forearm the succeeding ones at higher levels At birth all children are Mantoux negative In children of the hospital class at the age of two to three years 70% are Mantoux positive when they have been in contact with open cases of pulmonary tuber culosis whereas only 12 % are positive when they are non contact cases During school life 40% of these non reactors become infected with tuberculosis This is known as the primary infection with tuber culosis and in many cases gives rise to no symptoms of ill health Later when the primary focus has calcified it may be revealed by X ray examination About 20 to 30% of medical students and

nurses who begin work at hospitals are Mantoux negative in dicating that they have not been infected with tuberculosis and during their work in hospital the majority of the negative reactors become positive showing that infection has occurred At the time of conversion from Mantoux negative to Mantoux positive the individual may notice symptoms of ill health He or she may lose weight become easily tired run a slight temperature or erythema nodosum may appear Others show more definite evidence of pulmonary tuberculosis A shadow may be seen in the lungs on X ray examination or a pleural effusion may form It has been found in Great Britain by Daniels (16) that of 2 120 nurses who were Mantoux positive when first examined 43 or 2 % developed tuberculosis while working in hospital and of 450 negative reactors 27 or 6 % developed the disease

No nurse who is a negative reactor should be allowed to work in a sanatorium or look after cases of tuberculosis elsewhere Special care should be taken in general hospitals of nurses or students at the conversion period The chest should be X rayed and the nurse taken off duty for three months A second X ray should be taken at the end of six weeks Extra milk and eggs should be ordered If at any time during this observation period any of the symptoms of ill health mentioned above are noted the nurse or student should be warded a record of the temperature taken the sedimentation rate of the red cells determined and the sputum if any and stomach washings examined and cultured for tubercle bacilli

The following precautions should be taken by nurses looking after open cases of tuberculosis in a hospital or sanatorium —

1 Nurses must wear masks and gowns when carrying out any nursing treatment Masks are to be worn once only and after use placed in a special container for sterilisation Gowns must be changed daily

2 Sputum pots must contain 2 % lysol and must be changed twice a day They are to be soaked in 2 % lysol for twelve hours before emptying Gloves must be worn when handling sputum pots

3 All crockery and cutlery used by patients must be boiled in a special crockery boiler and then washed

4 All sheets pillow cases blankets pillows and mattresses must be put into a special container prior to sterilisation Nurses must wear masks and gowns when handling the bedding

5 Patients must wear a mask during examinations of the chest

6 Patients must use paper handkerchiefs These are to be placed in a special paper bag after use and burnt Handkerchiefs must not be kept under the pillow

7 All newspapers and books are to be placed after use in a dressing bin and then burnt

8 Any food not eaten by the patient must be put in the rubbish bin and burnt

9 On admission the patient s day and night clothes must be sterilised and then sent to the relatives or kept until the patient s discharge The patient shall wear hospital clothes which are subsequently sterilised with the bedding

In hospital unsuspected cases of pulmonary tuberculosis especially in surgical wards are a source of danger to nurses This has been shown by Stammers (17) The incidence is probably about 4 per 1 000 All patients admitted to hospital should have a chest examination by miniature radiography as soon after admission as possible

B C G Vaccination

For many years after the discovery of the tubercle bacillus by Koch in 1882 attempts were made to raise the resistance of the individual against the disease by means of a vaccine

The Bacille Calmette Guérin vaccine (B C G) is a stable living culture of bovine tubercle bacilli produced by Calmette and Guérin (18) in France in 1906 from an organism isolated from the milk of a tuberculous cow By subcultures over a period of thirteen years its virulence was reduced so that the bovine bacilli became avirulent to animals and man On injection into an animal the vaccine produced a localised nodular lesion with no dissemination of the disease It was found that virulent tubercle bacilli could be injected into the vaccinated animal without harmful results In 1921 Weill Hallé (19) first used B C G to vaccinate children in Paris and Irvine (20) states that by 1931 over a million children throughout the world had been so vaccinated In 1930 B C G fell into disrepute owing to a tragedy at Lubeck in Germany when after inoculating by mouth 271 infants 77 died of tuberculosis It was later proved that the vaccine used was a mixture of B C G and a virulent strain of living tubercle bacilli kept in the same incubator

Those responsible for this malpractice were imprisoned (21) Since then it is probable that no fatalities have followed the use of the vaccine

Attention has been directed to possible difficulties and disadvantages in the use of B C G by Wilson (22) Thus the vaccine should be used within fourteen days of its preparation It should be kept at a temperature of 3 to 6 C The virulence of the vaccine is not fixed so that if it is too virulent there may be harmful local reactions and if the virulence is too feeble inadequate protection will be afforded The vaccine must be injected into the superficial layer of the skin if it is given deeper long standing ulcers may form When used to vaccinate infants born of tuberculous mothers the infant must be taken away from the mother at birth or if taken away later the infant must be observed for six weeks after separation to make sure that it has not contracted tuberculosis After vaccination the infant must be kept away from a tuberculous parent for two to three months to allow immunity to develop The fact that the vaccination has taken is shown by the conversion of the Mantoux reaction from negative to positive and this occurs in over 85 / of cases vaccinated intradermally

Very satisfactory statistics have been published from Scandinavia Heimbeck (23) in Scandinavia found that a large proportion of student nurses became infected with tuberculosis early in their hospital training 50 / of the nurses being negative reactors on entering hospital Of the negative reactors 29 6 / developed tuberculosis but only 2 6 / of the positive reactors With the use of B C G to convert the negative to positive reactors the incidence of tuberculosis fell to 2 3 / in this group

Nordwall (24) in a training school for nurses in Stockholm found that before the use of B C G 40 / of the negative reactors developed tuberculosis during their training After vaccination with B C G only one nurse developed tuberculosis and that in a mild form Malmros (25) writing from Oerebro Sweden points out that conscientious tuberculin testin has perhaps been used to a greater extent in Scandinavia than elsewhere and it has been observed continuously that previously Mantoux negative adolescents fall ill with tuberculosis

Wallgren (26) who is largely responsible for the widespread use of B C G vaccination in Sweden says that revaccination is not

commonly done in that country The tuberculin sensitivity persists for three to ten years or more and affords protection during the most susceptible childhood ages

Hertzberg (27) of Oslo gives authoritative statistics of the results of B C G vaccination in Norway where such vaccination is now compulsory All those vaccinated were negative reactors and the majority became positive reactors two months later In many *cases the tuberculin reactions became stronger as time went on and* this is considered to be due to the supervention of a reaction due to the human type of bacillus *i e* to the vaccination allergy induced by the B C G is added a superinfection allergy due to contact with human tubercle bacilli as bovine tuberculosis has been eradicated from Norway Hertzberg found that protection is greatest when superinfection follows B C G vaccination at an interval of about four years

Rosenthal (28) and his colleagues in America record their experiences of thirteen years study of B C G They found that the application of the vaccine to all age groups is a safe and effective procedure They used the method of multiple puncture scarification

B C G vaccination is now being used in certain hospitals in England to give protection to the staff It should be offered to nurses or students who are negative on two consecutive tests made at an interval of two weeks to O T 1/100 The second test should be made with a different batch of O T from that used on the first occasion No vaccination must be given to an individual suffering from measles whooping cough eczema furunculosis or to one being inoculated against another infection such as diphtheria The B C G vaccination must be done immediately after the second negative Mantoux test The vaccine must be shaken well before use if there is more than a faint haziness or if clumps are observed in the fluid after shaking the ampoule should be discarded A glass *tuberculin syringe* fitted with a Luer needle No 25 or 26 (B W G) ¼ or ½ inch long is used for the injection The skin over the left deltoid is cleaned with spirit and 0 1 ml of the vaccine containing 0 5 mg per ml is injected strictly intradermally A wheal about 5 mm in diameter is produced No dressing need be applied In infants the injection may be made into the skin of the outer surface of the buttock Vaccination may also be performed by multiple puncture or by scarification A stronger vaccine is then required

containing 20 mg per ml This vaccine is not obtainable in this country For further information the reader is referred to the Ministry of Health Memo (29) After vaccination the nurse must be segregated from exposure to tuberculosis for six weeks The Mantoux test is then repeated and should now be positive to 1/100 O T If the test is negative vaccination should be repeated immediately and segregation again enforced for six weeks A local reaction with a small shallow ulcer about 10 mm in diameter may develop after three to six weeks which may take two months to heal The Mantoux test must be repeated every twelve months after successful vaccination and if it becomes negative re vaccination should be carried out

It is hoped that the results obtained in England will be as good as those in other countries but in every case of exposure to open tuberculosis there should be no relaxation of other safeguards against infection such as disinfection of sputum and avoidance of contact with droplet infection and with infected articles Pulmonary tuberculosis developing in adults over the age of forty accounts for a large number of cases in all countries The incidence in this group will not be affected by vaccination limited to children and young adults Further as stated above these infected adults are largely responsible for the development of pulmonary tuberculosis in young children In the programme already planned for the control of tuberculosis little attention has yet been paid to the problem of the re infection of adults over the age of forty

Mass Miniature Radiography

The use of mass miniature radiography for selected groups of ults such as factory workers and those in the services has aled an incidence of about 4 per 1 000 unsuspected cases of onary tuberculosis In some cases a shadow is seen in others vity The discovery of a shadow often causes unnecessary and despondency as frequently the disease is not active such a shadow is seen the patient should be admitted to l in order to try to decide whether or not the disease is active ictical difficulty is the long waiting period before a bed is and what the unfortunate individual is to do in the ile Those cases with a cavity are almost certainly active equate accommodation can be provided for the isolation

and treatment of the active and "open" cases mass radiography appears of little value either as a preventive or curative measure

Artificial Pneumothorax

Historical Carson (30) of Liverpool in 1822 first suggested that air might be admitted into the pleural cavity as a therapeutic measure in certain diseases of the lungs Cayley (31) in 1885 at the Middlesex Hospital London was responsible for the establishment of a *pneumothorax by pleural incision in a case of* intractable hæmoptysis Forlanini (32) in Italy must undoubtedly be regarded as the originator of the modern pneumothorax treatment by puncture for pulmonary tuberculosis his work dating back to 1882 Lillingston (33) in 1910 was one of the first to adopt this method in England

The Indications

Collapse of the lung by the introduction of air into the pleural space is chiefly of value in the treatment of pulmonary tuberculosis It is sometimes used for the relief of pain in dry pleurisy associated with pneumonia or with a peripherally situated bronchial carcinoma A diagnostic artificial pneumothorax may be helpful in determining whether a shadow shown by X ray examination is intrapulmonary in the chest wall or associated with the diaphragm or mediastinum

Pulmonary Tuberculosis There is a tendency now not to hasten to establish an artificial pneumothorax immediately pulmonary tuberculosis is diagnosed but to see first how the patient responds to bed rest combined with streptomycin and P A S therapy Artificial pneumothorax is of value in unilateral disease with or without thin walled cavities when dense fibrosis is not present It may also be employed in similar types of bilateral lesions either as a bilateral pneumothorax or as a unilateral pneumothorax combined with a phrenic crush operation on the other side and an artificial pneumoperitoneum In cases of hæmoptysis which do not respond to other methods of treatment the introduction of about 800 ml of air into the pleural space may sometimes control the bleeding It should only be attempted if it is reasonably certain which lung is the source of the hæmoptysis

III. CONTRA-INDICATIONS

An artificial pneumothorax should not be induced in miliary pulmonary tuberculosis nor in very toxic exudative lesions of the pneumonic or broncho pneumonic type In such cases the dyspnœa is increased by an artificial pneumothorax and the general condition of the patient rapidly deteriorates There is also a serious risk of the formation of a tuberculous empyema A satisfactory collapse is not usually obtained in chronic fibrotic lesions or with thick walled cavities Active tuberculous tracheo bronchitis especially when there is bronchial stenosis as shown by bronchoscopy is another contra indication Here again there is a risk of tuberculous empyema Tension cavities in the lungs resulting from partial bronchial obstruction so that air enters on inspiration and does not come out on expiration often become larger when the lung is collapsed by artificial pneumothorax

A cavity at the apex of the dorsal lobe is difficult to collapse by an artificial pneumothorax alone but an additional phrenic crush on the same side together with an artificial pneumoperitoneum has in our experience at times been successful In some cases a fair degree of collapse is obtained by the pneumothorax but the diseased portion of the lung is not adequately relaxed owing to adhesions Under these circumstances a thoracoscopic examination should not be made for four to six weeks as the adhesions if fine often stretch The thoracoscopist should always err on the side of caution in attempting to divide thick or awkwardly situated adhesions as serious sequelæ may result from hæmorrhage or empyema Despite what is said to the contrary those who have carried out practically every refill themselves on patients for many years will agree that many of their most satisfactory cases engaged in full work have what is known as a bad pneumothorax with dense adhesions and much pleural thickening Cavities with fluid levels however which do not close are an indication that the pneumothorax should be abandoned and some other form of collapse treatment attempted

As pointed out by Young (34) the greatest cause of failure of an artificial pneumothorax is lack of continuity in personnel The trouble has been that the technique seems so easy that it is carried out by inexperienced workers who do not understand either the proper selection of cases or the finer points in its conduct The

patient should always be screened immediately before the refill and the whole treatment lasting for several years should be carried out by one individual

The Method

The requirements are as follows —

Pneumothorax Apparatus There are several types in use The Lillingston and Pearson apparatus is that which we usually employ It has the advantage of simplicity, and can be used both for inserting air and also for removing it in cases of spontaneous pneumothorax

The arrangement of the pneumothorax apparatus can be seen by a reference to the diagram and a detailed description is not necessary (see Fig 43) A and B are glass bottles B being graduated in ml from 0 to 1 100 C is a glass manometer D is a four way glass connection E is a glass connection F and G are glass filters containing sterile wool H I J and K are glass tubes L M N O and P are rubber tubes Q is the needle 1, 2 3 and 4 are clips on the rubber tubes

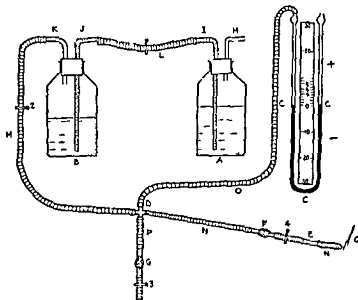


Fig 43 Diagrammatic representation of Modified Lillingston and Pearson Pneumothorax Apparatus The bottle B is graduated in ml from 0 to 1 100

Pneumothorax Needles The needle used for the primary induction is often referred to as the No 1 needle The form devised by Clive Riviere is commonly employed It consists of a blunt ended cannula with a thin edge for penetrating the pleura and a

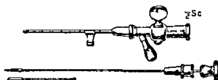


Fig 44 Clive Riviere Needle (No 1 Needle)

trocar for piercing the tissues of the chest wall The needle used for refills is of a different pattern it may be referred to as the No 2 needle The Saugman pattern is the one we prefer This is shorter than the No 1 needle and has a bevelled cutting edge like an ordinary hypodermic needle It is provided with a stilette for clearing purposes These needles are illustrated in the text (see Figs 44 and 45) The Morland needle has a solid point with a lateral opening near the tip It must be dry sterilised as a drop of moisture is very liable to cause blocking

A 2 ml syringe and hypodermic needle of sufficient length to reach through the chest wall to the pleura (This distance is rarely more than 3 cm)

Sterile 2 / procaine hydrochloride solution

Iodine spirit lamp and sterile towels

Some restorative such as brandy or adrenaline should be at hand

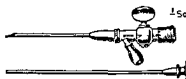


Fig 45 Saugman Needle (No 2 Needle) with Rubber Cap to act as a Stuffing box

Preparation of the Apparatus The glass bottles rubber tubes glass tubes and glass filters are all sterilised by boiling The needles are sterilised by placing them in spirit and subsequently flaming The manometer is filled to the zero point with water coloured with a little red ink This does not stain the glass and makes readings easy

The glass filters are filled with sterile cotton wool The tubes are placed in position Bottle A is filled with coloured solution of perchloride of mercury (1/1000) or with 1/100 carbolic acid The fluid is passed over to Bottle B, by blowing a little air into bottle A with a Higginson syringe attached to the tube H which starts the siphon action With the clips 1 2 and 3 open the fluid in the bottles can now be run from one to the other by raising or lowering one of them Half the fluid should be left in bottle B and half in A The apparatus should be examined to see that there are no leaks before use This can be done by closing clips 3 and 4 and opening clips 1 and 2 If now, bottle A is raised air will be driven from bottle B into the manometer and the fluid in limb C depressed The level in the manometer should remain constant as long as the bottle A is kept in the same position In the same way if bottle A is lowered the fluid in limb C will rise It can easily be ascertained that air can be driven through the pneumothorax needle by opening clips 1 2 and 4 and closing 3 If the end of the needle is now put under methylated spirit, having withdrawn the trocar and closed the side tap air will be seen bubbling from the end of the needle on raising bottle A

Preparation of the Patient The patient is in bed He should be given an aperient the night before the operation and the induction should not be made immediately after a meal Half an hour before the operation an injection of ¼ gr morphin sulph should be given in order to calm the patient and diminish any tendency to pleural shock If the pneumothorax is to be induced on the right side of the chest the patient lies on his back turned towards his left side with the right arm forward above the head which is kept low and a pillow is placed under the chest and left shoulder so as to widen the intercostal spaces The site of election for the first puncture is in the sixth space in the anterior axillary region The skin over this area is painted with iodine and sterile towels placed on the bed clothes and against the chest

The Operation The procaine solution is injected about 2 ml being used The skin over the intercostal space is held taut with two fingers of the left hand The needle is then inserted obliquely just in front of the fingers and a little procaine injected intradermally to cause a bleb After a short pause the needle is moved into a position at right angles to the chest wall and gradually

pushed through the intercostal space Procaine is slowly injected as the needle moves downwards to the pleura Care should be taken to inject the solution right down to the pleura as this probably plays a part in the prevention of pleural shock which may cause a fatal result

The patient must now be warned against coughing during the operation If the desire to cough is irresistible due warning must be given otherwise the sudden increase in the pleural pressure when the patient coughs with the pneumothorax needle in the pleural cavity would drive the fluid out of the manometer This can be prevented if warning is given by squeezing the tube N leading from the needle to the manometer or by closing the clip 4

The rubber tube N leading to the manometer is attached to the side arm of the No 1 needle Clips 1 2 and 3 are closed and clip 4 opened With the trocar in position the needle is pushed through the skin at the anæsthetised spot and through the intercostal muscles for about 1 cm or less if the patient is very thin It must be held firmly in the hand with the top of the trocar in the palm and not in the position of a pen between the fingers The trocar is now withdrawn and the stopcock turned so that the lumen of the needle is in communication with the tube leading to the manometer The cannula is now pushed on through the remainder of the intercostal muscle down to the pleura

When it is in contact with the pleura but not through it an oscillation will probably be observed in the manometer not directly synchronous with respiration and of small extent The cannula is now pushed through the pleura which can often be felt to give way with a snap Great care must be taken here not to penetrate the lung When the cannula is in the pleural cavity the fluid in the limb C of the manometer falls below zero and oscillates with respiration These oscillations correspond accurately with the respiratory movements falling with inspiration in the limb C of the manometer and rising with expiration A reading is now taken each centimetre of fall of fluid in the limb C corresponding with a change of pressure in the pleural cavity of 2 cm of water The oscillations should be at least 3 to 6 cm and are often as much as 10 cm Thus a reading of — 10 — 2 may be obtained giving a mean negative intrathoracic pressure of — 6 cm water The respiratory fluctuations are greater when the patient takes a very

deep breath but the reading should be taken with ordinary moderately deep breathing The needle may become blocked during its passage through the deeper parts of the chest wall in which case it can be cleared with the stilette which is supplied with the No 1 needle The visceral layer of the pleura may also come in contact with the end of the cannula and cause obstruction Letting in a small quantity of air by opening clips 1 and 2 is the best means of dealing with such a condition When it is certain that the end of the cannula lies free in the pleural cavity air may be admitted This is accomplished by opening the clips 1 and 2 when the air is sucked from the bottle B into the chest and fluid passes over from bottle A into bottle B to take its place When about 100 ml have thus passed in by suction the bottle A can be raised slightly and another 200 ml of air admitted The clips 1 and 2 are then closed and the manometric readings taken for inspiration and expiration with the patient breathing quietly and moderately deeply as before They will still be negative unless the air has been injected into a small loculated pleural space These readings may be — 8 — 1 The mean pressure would in this case be — 4 5 The pressure changes and the amount of air injected would then be recorded as follows — 6 300 ml — 4 5 *It is to be noted that with this pneumothorax* apparatus readings of intrapleural pressure cannot be taken while air is flowing in The clips 1 2 and 3 must be closed in order to obtain a reading The needle is now withdrawn from the chest and a little iodine again applied to the site of the puncture A collodion dressing is not usually required If there is a tendency to cough a large pad and a firm chest binder should be used in order to minimise extravasation of air The patient is kept in bed at rest for the first month of pneumothorax treatment

Refills The same apparatus is required as for the initial pneumothorax operation but a No 2 needle (Saugman) is used The patient is prepared as before but it is not usually necessary to give a preliminary injection of morphine unless he is very nervous It is wise however not to omit the anæsthetisation of the pleura with procaine as death may follow from pleural shock in refills as well as during the primary induction A spot close to that used for the induction is chosen The rubber tube leading to the manometer is attached to the side arm of the No 2 needle with the stilette slightly withdrawn so that it does not protrude beyond the point The

pneumothorax apparatus has already been prepared with the fluid in bottle B up to the 100 mark the remainder being in bottle A The clips 1 2 and 3 are closed The needle is now pushed straight through the chest wall at the anæsthetised spot into the pleural cavity Directly it is felt to enter the stilette is withdrawn and the stopcock closed so that the lumen of the needle is in communication with the manometer The fluid in the manometer will now oscillate The pointed needle is used as the air introduced at the primary induction (on the previous day) will prevent the lung being injured

The reading is then taken This will be found to be lower than the final reading at the previous injection as some of the air introduced has been absorbed Clips 1 and 2 are now opened and air allowed to enter the pleural cavity After about 200 ml have passed in the clips should be closed and a reading taken to make sure that the needle is in the pleural cavity and that the pressure is not rising too quickly About 400 or 500 ml will probably be enough for the first refill in order to reduce the negative pressure in the pleural cavity by about 2 or 3 cm of water Thus the readings at the first refill may be — 5 5 400 ml — 2 If there are many adhesions and the lung is not collapsing well a smaller quantity of air should be required to reduce the negative pressure to a similar degree The needle is then withdrawn

The spacing of refills and the determination of the correct amount of air to be injected are matters of extreme importance and are comparable with the administration of a vaccine In neither case is it a matter of routine in each the personal factor of the patient is of paramount importance Usually the second refill is given two days after the first and the third three days after the second The interval can then be gradually lengthened to a week ten days a fortnight three weeks and a month High intrapleural pressures should be avoided as they cause mediastinal displacement possibly tend to promote pleural effusion and seem to have a direct relation to loss of weight on the part of the patient

If there is a rise of temperature after the initial induction the first refill should not be given the next day The temperature should be allowed to subside first A rise of temperature just before a refill is due probably indicates that the refills are being given at too long intervals and that the lung is starting to re expand A rise of temperature after a refill is sometimes an indication that too

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much air has been injected An X ray should be taken at the end of the second week to determine the degree of pulmonary collapse and the subsequent treatment should be subjected to X ray control in order to determine what degree of collapse is necessary to produce the optimum effect as judged by temperature pulse and body weight and also so that the range of intrapleural pressure which produces such collapse can be ascertained It is not usually necessary to use a positive pressure to produce satisfactory collapse

Some Difficulties and Complications The manometer has been described as the heart of the pneumothorax apparatus It is certainly of vital importance not for the actual introduction of the air but for its safe introduction

The character of its oscillations affords the only reliable guide as to whether the end of the needle lies free in the intrapleural space These typical movements have already been described

Certain abnormal events may occur in which case no air must be admitted into the chest —

A negative pressure may be obtained on inserting the needle into the chest but the fluid in the manometer may show this as a stationary negative pressure without any respiratory excursions This indicates that the needle has been in the pleural cavity but is now either blocked or in the lung The negative pressure may increase for a little with each inspiration without any oscillations occurring if the needle is in the lung The pressure may be slightly positive during expiration at the primary induction and negative during inspiration indicating that the needle is in a bronchus or small cavity in the lung The manometer may show a positive pressure which rises slowly or rapidly indicating that the needle is in a blood vessel Blood may then be seen passing through the glass connection E in the rubber tube attached to the needle The pressure may be negative but show irregular respiratory oscillations if the needle is in a pleural cavity much loculated by adhesions From such examples as these it will be realised how important it is to be certain that the manometer indicates that the needle is in the pleural cavity before any air is admitted A refill may be very difficult if the lung has expanded and is touching the chest wall In such a case the patient should expire deeply and hold the breath in expiration as the needle is inserted through the pleura In this way the pleural space can often be found

Complications *Pleural shock* may occur especially if the patient is very nervous and if the pleura is inflamed and not anæsthetised Some authorities deny the existence of pleural shock but their evidence is not conclusive

Surgical emphysema may develop within a few hours of the injection of air either as the result of coughing or of injury to the lung or because the air has been injected extrapleurally between the pleura and the intrathoracic fascia If a lung which is bound down to the chest wall by adherent pleura is punctured surgical emphysema may develop without the introduction of any air

Air Embolus This may occur from introduction of air into a vessel especially a pulmonary vein in which the pressure is negative

Effusion This is especially liable to occur if adhesions are present and if high positive intrapleural pressures are used About 50% of pneumothorax cases develop an effusion in the pleura during some time of their treatment This may be *Small* giving rise to little constitutional disturbance and often unrecognised except by X rays *Large* causing constitutional disturbance with pyrexia The fluid is serous rich in cells and frequently contains tubercle bacilli It indicates an active tuberculous pleurisy *Infective* often accompanied by rigors This is pyogenic in origin

In any case pleural adhesions are likely to occur both at the apex and the base and it is improbable that a satisfactory collapse will be obtained afterwards unless the fluid is repeatedly removed as soon as it forms and is replaced by air The apical adhesions form because the fluid runs up to the top of the pleural space when the patient lies down

Atelectasis In some cases of artificial pneumothorax especially after division of adhesions a portion of the collapsed lung becomes atelectatic Maher Loughnan (35) has drawn attention to this complication and its frequent association with pleural effusion and considers it an important cause of relapse in cases of pulmonary tuberculosis treated by unilateral artificial pneumothorax We have treated several cases of this type and found that by alternately allowing the lung nearly to expand and subsequently collapsing it again it is often possible to effect re aeration of the atelectatic portion of lung It is also helpful to tilt the foot of the bed on 8 inch blocks

Duration of Treatment

It is always difficult to decide when the pneumothorax should be abandoned in cases which are maintaining good health A refill every three to six weeks or at longer intervals is a small price to pay for health and there is no means of knowing whether the disease will become active again when the lung is allowed to expand Further when the pneumothorax has been maintained for several years there is a possibility that as the lung expands it will displace the mediastinum to the affected side and cause respiratory difficulties As a working rule it may be said that the pneumothorax should never be abandoned in less than two years that five years is an average life for a pneumothorax and if it is continued for over seven years it is probably wiser to go on with it indefinitely

A pneumothorax had been maintained for six years in one of our patients in whom there had originally been a cavity in the right upper zone The right lung appeared well controlled and the patient was in good health She wished to abandon the pneumothorax She came into hospital and the lung was allowed to expand with diminishing refills under streptomycin cover Three and a half months later she did not feel well and on X ray examination we found an oval shadow (5 5 × 2 5 cm) of uniform density in the right upper zone just above the right interlobar fissure The disease was once more active

Artificial Pneumoperitoneum

Historical By introducing air into the peritoneal space the diaphragm if free from adhesions can be raised 3 or 4 or more inches with a corresponding diminution in the volume of the lungs (see Fig 46) The rise is usually greater on one side than the other especially when the pneumoperitoneum is combined with a phrenic crush A lateral view of the chest however may show that only a portion of the diaphragm is elevated

The method was introduced by Vajda (36) in Hungary in 1933 for the treatment of bilateral pulmonary tuberculosis and in 1934 Banyai (37) *combined pneumoperitoneum* with phrenic paralysis to obtain a more effective pulmonary collapse During the last decade this method has been more extensively employed and representative articles published on the subject in America

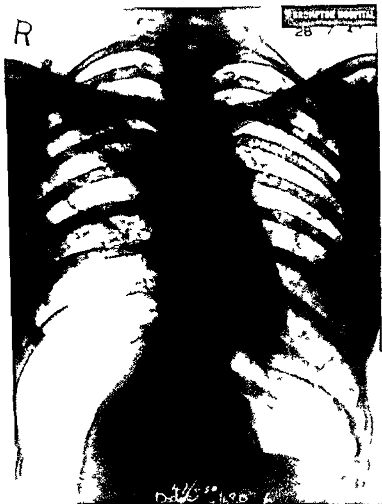


Fig. 46 Pneumoperitoneum and left phrenic crush

[To face p 62

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include those by Rilance and Warring (38) Anderson and Winn (39) Crow and Whelchel (40) Mitchell and his colleagues (41) Gilmore (42) and Murphy (43) In Great Britain reference may be made to the writings of Keers (44) and Edwards and Logan (45)

The Indications Pneumoperitoneum is of value in the treatment of basal mid zone and apical lesions Anderson and Winn (39) found that in a series of 113 cavities 58 6 / of apical 57 7 / of mid zone and 58 8 / of basal ones were closed by pneumoperitoneum treatment In 91 cases with a positive sputum 56 / became negative with the treatment We have employed it with satisfactory results in various types of pulmonary tuberculosis including apical lesions in which an artificial pneumothorax was impossible owing to adhesions in cases of pneumothorax in which adhesions were present to try and divide which would have been dangerous and in bilateral lesions In the majority of cases a phrenic crush should be performed on the side on which the disease is most advanced and streptomycin or streptomycin combined with P A S (para aminosalicylic acid) can with advantage be administered simultaneously Pneumoperitoneum is helpful in controlling small areas of disease in one lung in preparation for a thoracoplastic operation on the more affected side A dorsal lobe cavity which does not collapse in response to an artificial pneumothorax combined with a phrenic crush may sometimes be successfully closed by a pneumoperitoneum Pneumoperitoneum is also of help in the emergency treatment of a severe hæmoptysis in which case it is not necessary to know from which side the bleeding is occurring We usually prefer to establish the pneumoperitoneum three to four weeks prior to the phrenic crush but with an apical dorsal lobe cavity the pneumoperitoneum and phrenic crush should be instituted at about the same time

As Mitchell and his co workers (41) state with almost no exceptions pneumoperitoneum can be abandoned and re established at will as the space does not become lost owing to an adhesive peritonitis

The Method The requirements are the same as those for artificial pneumothorax but a Saugman refill needle is used both for the induction and for refills The fluid in the bottles of the pneumothorax apparatus is adjusted for the induction and for refills with the fluid in bottle B at the 100 mark the remainder being in bottle

a confirmatory sign that the needle is in the peritoneal space The needle is now withdrawn and the puncture sealed with gauze and collodion The patient should lie flat for twenty four hours after the induction

The first refill is given the next day the second refill four days later and the third refill five days after that Subsequently refills are given every seven days The amount of air required is usually 800 to 1 000 ml and the pressures are found to be positive such as +6 1 000 ml +12 cm water It is unwise to raise the final pressure above a mean of +1° At each refill a few bubbles of air should be sucked back into the syringe on injecting the local anæsthetic to indicate that the needle is in the peritoneal space Some writers advice the patient to wear an abdominal belt after the establishment of the pneumoperitoneum but we have not found this necessary unless the abdominal muscles are very lax The electrocardiogram shows changes due to rotation of the heart with displacement of the apex upwards and outwards T becomes flattened T_3 inverted and Q_3 is apparent

Some Difficulties and Complications The chief difficulty is to be sure that the needle is in the peritoneal space It is probably not uncommon for the intestine to be perforated but usually no harm results and if the air is injected into the colon it is quickly passed per rectum Perforation of the stomach may give rise to more serious symptoms In one of our cases a pneumoperitoneum was induced at 11 a m The readings recorded were 0 500 ml +4 At 7 45 p m there was a sudden attack of dyspnœa with severe collapse The patient was pale sweating the pulse rapid and frequent and severe pain was complained of in the left subcostal region and top of the left shoulder The temperature rose to 100 4 F falling to normal during the next three days There was no vomiting and no abdominal rigidity The next day the patient was improving although he still looked pale and ill An X ray examination showed air below each diaphragm It was thought that there had been a small perforation of the stomach which rapidly sealed over Other severe complications include intra abdominal hæmorrhage from perforation of mesenteric vessels which may cause death Air embolism with the rapid onset of convulsions is not necessarily fatal Peritoneal effusion forms in about 4 / of cases and is probably present in a larger proportion

A If the patient is nervous phenobarbitone gr 2 should be given by mouth three quarters of an hour *before* the *induction* The patient lies on his back in bed with his head resting on a pillow The initial puncture should be made on the left side 1½ inches below the subcostal margin at the outer edge of the rectus muscle Some authorities such as Gilmore (42) prefer a point 2 to 3 inches to the left about an inch below the umbilicus If the puncture is made on the right side *there is danger of injuring the liver or gall bladder Proximity to an abdominal scar* must always be avoided owing to the risk of abdominal adhesions beneath the scar The skin is painted with iodine and 2 ml of 2% procaine are injected through the skin and subcutaneous tissues down to the peritoneum The Saugman needle with stilette inserted but not passed through to the needle point is *attached to the rubber tube N of the pneumo thorax* apparatus *and slowly pushed through the abdominal wall* the clips 1 2 and 3 on the rubber tubes of the apparatus being closed The patient must raise his head and shoulders off the pillow not resting on his elbows while the needle is being inserted in order to render the abdominal muscles tense The peritoneum can usually be felt and there may be some pain as the needle penetrates it *The patient now rests his head again on the pillow* When the *end of the needle is in the peritoneal space it will be found* that the stilette can be passed through to the hilt the end of the stilette now projecting just beyond the needle tip This is not possible when the end of the needle is in the tissues of the abdominal wall The common mistake is not to insert the needle far enough The stilette is now withdrawn the stopcock on the needle closed so that the *lumen communicates* with the manometer Usually no swing is seen on the *manometer as the patient breathes About 40 ml* of air are run in by opening the clips 1 and 2 The initial pressure is now taken by closing clip 2 This is usually 0 and on breathing a slight manometric swing can be seen the pressure rising with inspiration and falling with expiration In some cases there is a negative pressure if the needle is inserted just underneath the *diaphragm After running in about 800 ml* of air the pressure *may rise to +6 +- cm water or it may still remain at zero* The liver dulness usually *disappears as the air is introduced often* before a pressure reading can be obtained Flicking the abdominal wall in the right iliac region may cause a manometric swing and this is

in a report to the Medical Research Council in 1950 tried the effect of varying the administration of streptomycin in four ways In one group of cases 0 5 g of streptomycin was injected six hourly in alternate weeks in the second group 0 5 g was given six hourly in alternate months in the third group 0 25 g was administered six hourly without intermission and in the last group 1 g of streptomycin was injected once a day without intermission The total course in each group lasted for six months The cases chosen were those of acute progressive bilateral lesions of presumably recent origin in the age group 15 to 30 years No delay in the emergence of streptomycin resistant bacilli was found in any group and from the clinical point of view there did not appear to be any difference between the continuous or intermittent forms of treatment The method of choice appears to be to give 1 g of dihydro streptomycin in a single dose daily and this causes least inconvenience to the patient

Para aminosalicylic Acid (P A S)

Bernheim (49) in 1940 showed that the addition of sodium salicylate to a phosphate buffer solution of *p*H 6 7 containing living tubercle bacilli stimulated the rate of oxygen consumption Lehmann (50) in 1946 found that only pathogenic tubercle bacilli showed an increased oxygen consumption in the presence of salicylates or benzoates After testing over 60 allied compounds Lehmann (51) subsequently found that para aminosalicylic acid was the most active in producing a bacteriostatic effect upon the tubercle bacillus Favourable results were soon obtained with the use of P A S on animals infected with tubercle bacilli and Lehmann (51) in 1946 first employed the drug clinically to treat tuberculous abscesses following thoracoplasty which had remained unchanged for several months before being so treated Daily local injections of a 10 / neutral solution of P A S were given with favourable results He also administered the drug orally in doses of 10 to 15 g daily in courses of eight days There was a rapid improvement in the patient s condition with fall in temperature and decrease in the sedimentation rate of the red cells Vallentin (5?) in 1946 used P A S in the treatment of pulmonary tuberculosis tuberculous empyema miliary tuberculosis and tuberculous meningitis No favourable effect was noted in the case of miliary tuberculosis or

although in insufficient amount to be detected clinically Cohen (46) has pointed out that a tuberculous ascites may develop in cases of pneumoperitoneum even when the pulmonary lesion is probably arrested or healed This is very liable to be followed by adhesive peritonitis Pneumoperitoneum should be abandoned immediately ascites is detected In cases of myocardial weakness the establishment of a pneumoperitoneum may precipitate an attack of congestive heart failure

In some cases the digestion and appetite are seriously impaired and nausea vomiting or pain may necessitate the abandonment of *the pneumoperitoneum Pain may be referred to one or other* shoulder if there are diaphragmatic adhesions and we have noted such pain to disappear after the phrenic crush has paralysed the affected side of the diaphragm Air may pass from the peritoneum into the mediastinum into the pleura into the subcutaneous tissues or along the inguinal canal into the scrotum forming there a pneumocele In other cases a small umbilical hernia may appear Rilance and Warring (38) in 1944 noted 7 cases of acute appendicitis in 101 cases undergoing pneumoperitoneum treatment an incidence eleven times greater than in other sanatorium patients

Streptomycin and Para aminosalicylic Acid in the Treatment of Pulmonary Tuberculosis

Streptomycin is still being used in the treatment of pulmonary tuberculosis The two main drawbacks are the possibility of vestibular nerve involvement with giddiness which often persists for long periods and may be permanent and the appearance of drug resistant tubercle bacilli which renders further administration of streptomycin useless With smaller doses of streptomycin and possibly owing to greater purification of the substance giddiness is a less frequent complication We have treated a considerable number of patients suffering from pulmonary tuberculosis with streptomycin and have only had two cases of severe giddiness The drug should always be discontinued directly any evidence of vestibular involvement appears It was hown in a report to the Medical Research Council in 1948 (47) that resistant organisms are generally found within two months of beginning treatment Even before treatment a minute number of streptomycin resistant tubercle bacilli can be found in the sputum *Bignall and his colleagues (48)*

in a report to the Medical Research Council in 1950 tried the effect of varying the administration of streptomycin in four ways In one group of cases 0 5 g of streptomycin was injected six hourly in alternate weeks in the second group 0 5 g was given six hourly in alternate months in the third group 0 25 g was administered six hourly without intermission and in the last group 1 g of *streptomycin was injected once a day without intermission* The total course in each group lasted for six months The cases chosen were those of acute progressive bilateral lesions of presumably recent origin in the age group 15 to 30 years No delay in the emergence of streptomycin resistant bacilli was found in any group and from the clinical point of view there did not appear to be any difference between the continuous or intermittent forms of treatment The method of choice appears to be to give 1 g of dihydro streptomycin in a single dose daily and this causes least inconvenience to the patient

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tuberculous meningitis In exudative pulmonary tuberculosis there was a rapid clinical and radiological improvement with loss of tubercle bacilli from the sputum but on discontinuing the treatment relapse frequently occurred In 1947 Dempsey and Logg (53) reported similar encouraging results in cases of pulmonary tuberculosis in Britain and they noted a reduction in the size of cavities during treatment Erdei (54) in 1948 commented on the very great improvement in the patients general condition which was usually noted about the third day of P A S treatment In 1949 Joules and Nassau (55) treated 10 cases of extensive bilateral pulmonary tuberculosis and although some benefit was noted in half the cases they concluded that in none of the patients was the prognosis materially altered for the better as the effect of the drug was only temporary Nagley and Logg (56) considered that the main value of P A S is in the treatment of the acute exudative type of pulmonary lesion especially in rendering such patients fit for collapse therapy It has the advantage over streptomycin that it does not produce P A S resistant strains of tubercle bacilli except possibly after prolonged treatment Thus Carstensen and Andersen (57) reported from Sweden that resistance to P A S develops in over 50 % of cases treated for four-five months Graessle and Pietrowski (58) in America produced evidence that the combined use of P A S in relatively low concentration with streptomycin (approximately 0 5 micrograms to 1 unit) prevents or greatly retards the *in vitro* development of streptomycin resistant tubercle bacilli It was shown in a report to the Medical Research Council in 1949 (59) that the incidence of streptomycin resistant bacilli can be considerably lowered by the combined use of streptomycin and para aminosalicylic acid This is the method which is being used more extensively at the present time

Method of Administration P A S is given as the sodium salt by mouth in doses varying from 12-24 g every twenty four hours It can be administered as sugar coated tablets (Paramisan sodium) each containing 0 33 or 0 5 g of the sodium salt as sugar coated granules (Paramisan sodium) containing 33⅓% by weight of the sodium salt as enteric coated tablets (Bactylan) containing 0 69 g of the sodium salt or as cachets (Paramisan sodium) each containing 1 5 g Alternatively it can be prescribed in a mixture which however flavoured has a bitter and unpleasant taste The

prescription which we use as an alternative to the tablets is Sod para aminosalicyl gr 84 (3 g) saccharin gr [1] emulsio cassiæ m 5 emulsio chloroform m 15 syrup simplic m 60 aq ad fl oz 1 This is taken after a little food every two and a half hours from 9 a m to 9 30 p m (= 18 g) for six days a week It should be freshly prepared each week In some patients P A S causes nausea vomiting or diarrhœa but this often passes off in a few days An alkali taken just before each dose will often relieve the gastric symptoms and the diarrhœa may be checked by a bismuth chalk and opium mixture If these measures fail the dose of P A S should be halved Some of our patients have been unable to take the drug the sight of the bottle or of the tablets or the thought of the next dose provoking intolerable nausea

Other side effects are uncommon but they include a skin irritation or rash drug fever hæmaturia or albuminuria and hypopro thrombinæmia In cases in which there is albuminuria or hæmaturia the drug should be stopped and the urine rendered alkaline By keeping the urine alkaline it may then be possible to continue the course as soon as the urine is clear

Madigan and his co workers (60) showed that hypopro thrombinæmia can be prevented or cured by the daily oral adminis tration of 10 mg of synthetic vitamin K Cayley (61) drew attention to the depression in the serum potassium level which may occur during P A S treatment This leads to paralytic limb lesions and disturbances of cardiac rhythm It seems probable that the fall in blood potassium is due to the sodium in the salt administered as suggested by Nagley (62) A calcium salt (Paramisan calcium) is also available in powder form for preparing solutions and in cachets This would probably avoid this complication

P A S is rapidly excreted in the urine and in order to obtain the best results a blood concentration of 5 to 10 mg per 100 *ml* should be maintained The P A S treatment may be continued effectively for periods up to 5 or 6 months

Conclusion Neither streptomycin nor P A S can be regarded as the long awaited cure of pulmonary tuberculosis although striking and notable results are achieved in many instances especially in the recent exudative types of lesion and in tracheo bronchial tuberculosis We have not seen tuberculous cavities of any size close during the treatment P A S has the advantage that

it can be safely administered at home while the patient is awaiting admission to hospital or sanatorium

Bronchography

It is not possible to visualise clearly the outline either of the bronchi and their ramifications or of pulmonary cavities by means of a direct X ray examination If however some substance opaque to the rays is introduced into the bronchi their configuration is clearly revealed

Lipiodol or Neo Hydriol (ol iodisat B P Add) is a compound of iodine in poppyseed oil which contains 40% iodine It is a transparent oil of a light brown colour which is opaque to the X rays and so heavy that it sinks in water On prolonged exposure to air it darkens and should not then be used for injection Sicard and Forestier (63) in 1922 injected Lipiodol into the trachea in man using a cannula introduced into the glottis with the aid of a laryngoscope or else passing it through a curved needle inserted through the crico thyroid membrane

The Indications X ray photographs of the lungs after intra tracheal injection of Lipiodol afford a graphic representation of the position of the trachea and main bronchi of cavities in the lung connected with patent bronchi of bronchial and pleural fistulæ and of the relative permeability of the bronchial fields in different portions of the lungs

Thus it will show displacement of the trachea and bronchi due to pulmonary *fibrosis or other causes the presence of bronchiectasis* in its various stages and whether or not the lumen of the bronchi is obliterated by obstruction within or without such as that caused by a foreign body new growth or fibrosis Lipiodol injections are of value in suspected cases of bronchiectasis to determine whether there is a cavity present and if so the extent of lung involved They also show the condition of the apparently sound lung A lobectomy for bronchiectasis would generally be contra indicated if the Lipiodol injection showed that the opposite lung was also the seat of bronchiectatic changes

The X ray appearance of the lung in bronchiectasis after Lipiodol injection is shown in the accompanying plate (see Fig 47)

Methods Iodised oil (Neo Hydriol Lipiodol etc) may be introduced into the bronchial tree to render it radio opaque by



Fig 47 Radiogram of lung after injection of Lipiodol showing normal and dilated bronchi

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one of the three following routes — (1) Through the crico-thyroid membrane (2) Over the back of the tongue (3) Via the nose

With practice any of these methods can give satisfactory results in a high proportion of cases but the most consistently reliable is the crico-thyroid route It is easier to fill the upper lobes by this method a minimum of surface anæsthetic is required and the whole procedure can be completed in fifteen minutes It should be avoided however in patients having large quantities of purulent sputum owing to the risk of subsequent cellulitis of the neck. Whichever method is to be used a few days preliminary postural drainage is advisable to empty the bronchi of sputum as far as possible In addition the patient should be tested for sensitivity to iodine and cocaine derivatives Ten grains of potassium iodide by mouth t.d s should be given for three days to see if iodism results as judged by lachrymation nasal catarrh and a skin rash In patients who are iodine sensitive an erythematous rash may appear one to three days after the operation or an urticarial rash about the tenth day and fatalities have been reported If therefore the patient is sensitive it is wiser not to proceed with the investigation.

After a satisfactory iodide test two minims of 2 / amethocaine hydrochloride or 5 / cocaine solution are injected subcutaneously idiosyncrasy to the drug being shown by faintness pallor dyspnœa, tachycardia and tremors Unfortunately a negative test is not a guarantee of insensitivity but if such symptoms do arise the investigation should not be continued except under a general anæsthetic

Premedication is seldom required but an apprehensive subject should be given morphine sulphate gr ¼ by injection or gr 4 of hexobarbitone orally half an hour beforehand

The operation should be carried out in the X ray department with the patient lying on a table capable of being tilted in either direction

Crico thyroid Route

Apparatus Minim syringe with fine needle (No 17 hypodermic) Minim syringe with stouter needle (No 12 hypodermic)

Lipiodol syringe This should be strong as considerable pressure is required to force the oil through the needle We use a 25 ml syringe with three finger rings the needle being straight (No 15 S W G ¾ inch long) short bevelled with a bayonet *fitting to the* syringe

right the head being kept extended A further 3 to 4 ml of oil are injected and another half minute allowed for this to fill the middle lobe He is then replaced in the semi recumbent position but lying completely on his right side a further 4 to 5 ml of oil are introduced and the needle is then withdrawn A few seconds are *allowed for the oil to reach the upper lobe bronchus* All pillows except one for the head are now removed The patient s feet are raised and the head lowered by tilting the adjustable table in order to fill the upper lobe The pectoral branch is best filled by *turning* him for a few seconds into the prone position X ray films in inspiration are now taken in antero-posterior and lateral positions with the patient recumbent and then repeated with him erect The needle is then reintroduced through the crico thyroid membrane and the left lung filled in exactly the same way but with the patient turned always slightly to the left It should be remembered that *on the left side as the upper* lobe bronchus *comes off rather more* anteriorly the patient should be inclined somewhat more to the prone position during its filling Films are then taken in the antero-posterior and left posterior oblique positions If only the left side has been filled a left lateral film can also be taken The procedure is a simple application of a knowledge of the anatomy of the bronchial tree as depicted in Fig 48

After completion of the operation postural drainage is advisable to remove most of the injected oil and the patient is not allowed to eat or drink until the anaesthetic effect has worn off

ORAL ROUTE

Apparatus 2 ml syringe
Curved metal cannula (Record fitting)
Lipiodol syringe
Curved metal cannula with bayonet fitting
Conical measuring cylinder graduated in ml
Sterile 2 / amethocaine hydrochloride solution
Adrenaline hydrochloride solution 1 in 1 000
Neo-Hydriol
Gauze swabs
Sodium thiopentone oxygen and carbon dioxide for emergency use

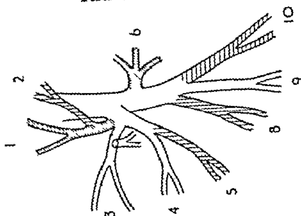
The Operation The *patient sits on an armless chair facing the*

Sterile ½% procaine hydrochloride solution
Sterile 2% amethocaine hydrochloride solution
Neo Hydriol
Iodine and wool

Sodium thiopentone or other rapidly acting intravenous barbiturate should always be immediately available along with oxygen and carbon dioxide in the event of idiosyncrasy to the surface anæsthetic manifesting itself

The Operation The patient lies in a semi recumbent position on the adjustable table supported by three or four pillows beneath the shoulders the head and neck being straight and extended as far as possible over a sandbag The skin of the neck is painted with iodine the crico thyroid membrane identified and the skin subcutaneous tissues and crico thyroid membrane anæsthetised by injecting ½ to 1 ml of ½% procaine through the fine needle Next using the stouter needle m 7 to 10 of 2% amethocaine hydrochloride are injected *rapidly into the larynx through the same puncture* wound A few bubbles of air should be sucked back into the syringe before the injection is made to ensure that the point of the needle is in the lumen of the larynx and the injection should be made at the end of expiration The needle is withdrawn immediately after the injection which will produce a bout of coughing The patient is now told to sit up and cough until he no longer feels a desire to do so He should spit out and not swallow the excess of local anæsthetic When the coughing has subsided he is warned that any cough after injection of the iodised oil will spoil the subsequent X ray films He now resumes the semi recumbent position with the head well extended as before The Neo Hydriol is poured into the special syringe and the needle introduced through the crico thyroid membrane while the larynx is steadied with the operator s free hand On withdrawing the piston a bubble of air will indicate that the needle is in the correct position and this should be checked at frequent intervals during the injection especially after a change in the patient s position He is now turned slightly to the right side and instructed to breathe quietly through the mouth Four or five ml of iodised oil are then injected and about half a minute is allowed for this to run down to the bronchi of the lower and dorsal lobes With the needle still *in situ* the patient is carefully sat up and inclined forwards and to the

operator with his tongue protruded and protected from the lower incisors by means of a gauze swab Four ml of 2% amethocaine and 1 ml. of 1 in 1 000 adrenaline are now poured into the conical measuring cylinder and this total amount should not be exceeded One ml of this mixture is drawn into the 2 ml syringe and injected rapidly over the back of the tongue by means of the curved cannula This will cause the patient to cough and so disseminate the local anæsthetic He should spit out any excess and avoid swallowing it A further 1 ml is then injected into each pyriform fossa in turn in the same way Alternatively a pledget of wool twisted on the end of a malleable probe and soaked in the anæsthetic solution may be held gently in each pyriform fossa until the desire to cough and retch has subsided Some minutes are allowed for the anæsthetic to take full effect and the index finger of the left hand is then hooked gently over the back of the epiglottis and base of the tongue If surface analgesia is adequate this should cause no distress and the superior aperture of the larynx can be identified A further 1 ml of the anæsthetic solution is then injected down the trachea between the vocal cords by guiding the curved cannula along the side of the left index finger and into the upper part of the larynx. Some operators prefer to use a laryngeal mirror in order to see the exact position of the cannula rather than to rely on palpation When coughing has subsided the patient s tongue is held well forward by means of a gauze swab wrapped round its anterior third, as in this position it is almost impossible to swallow The patient is instructed to breathe quietly through his mouth and to lean towards the right but keeping his head upright By means of the Lipiodol syringe and its attached cannula the iodised oil is then dripped steadily on to the back of the tongue Provided the patient does not swallow it will flow round either side of the epiglottis and down the larynx While the first 5 ml are being injected the patient is inclined backwards and to the right to fill the right lower lobe during the injection of the next 4 or 5 ml he should lean forward and to the right to fill the middle lobe and as soon as the third 5 ml have been injected he should lie on the adjustable X ray table and be postured for filling the branches of the upper lobe as described for the crico-thyroid method When films have been taken of the right side the procedure is repeated for the left lung but with the patient inclined towards the left side throughout



UPPER LOBE
1 Apical bronchus
2 Posterior bronchus
3 Anterior bronchus

MIDDLE LOBE
4 Lateral br
5 Medial br

LINGULA
4 Superior br
5 Inferior br

LOWER LOBE
6 Apical bronchus ← → 6 ditto
7 Medial basal (cardiac) br
8 Ant basal br — 8 ditto
9 Lateral — 9 ditto
10 Post — 10 ditto

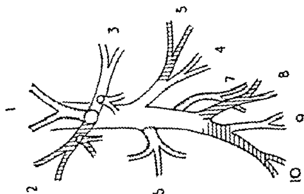


Fig 48 Diagram of scheme of nomenclature of the segmental bronchi (Brock *Thorax* 1950 5 222)

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Nasal Route

The Operation The patient sits facing the operator with his head fully extended his tongue protruded and held by means of a gauze swab A short piece of sterile thin rubber tubing 1½ to 2 inches long is attached to the 2 ml syringe and inserted for a distance of 1 to 1½ inches into the more patent nostril beneath the inferior turbinate The same anæsthetic solution is used as for the oral route He is then told to inhale deeply through the nose and at the beginning of inspiration 1 ml of the surface anæsthetic is injected through the rubber tubing As the solution reaches the pharynx and larynx he will cough and should spit out the excess A further 1 ml is then injected in the same way and this will usually be found to produce satisfactory anæsthesia after waiting for some minutes The piece of thin rubber tubing or another similar piece is then attached to the nozzle of the lipiodol syringe and the iodised oil is injected steadily through this into the anæsthetised nostril With the patient breathing through his nose head extended and tongue held well forward to prevent swallowing the oil will flow down the pillars of the fauces fill up the pyriform fossæ and then overflow and spill down the larynx During the injection of the oil posturing is the same as already described for the oral route

Bronchography in Children

Children under the age of ten will usually require a general anæsthetic of sufficient depth to abolish the cough reflex Pre medication is advisable and Omnopon gr 1/30 per stone of body weight together with atropine sulphate gr 1/150 to 1/100 is very satisfactory Anæsthesia is induced with cyclopropane and an endotracheal tube is inserted via the mouth with the aid of a laryngoscope The anæsthetic is then continued with gas oxygen and ether Ether can be recommended because of its broncho dilator action A T piece is attached to the outer end of the endo tracheal tube and one orifice is corked during the induction When anæsthesia is fully established this cork is removed and the bronchial tubes are aspirated through a gum elastic catheter connected to a sucker To the Lipiodol syringe is attached a soft nasal catheter which has a mark on it indicating the length of the endotracheal tube After aspiration of the bronchi this catheter is inserted down

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The anatomical distribution of the bronchial tree should be borne in mind in all these operations as depicted by Brock (64) (see Fig 48)

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CHAPTER X

ANTITHYROID SUBSTANCES

Iodine

Previous to the publications of Plummer and Boothby (1) in 1924 there was considerable difference of opinion concerning the advisability of administering iodine to patients suffering from Graves disease In this year Plummer showed in a series of 400 cases that large doses of Lugol s solution 10 to 15 m once or twice daily given as a pre operative measure caused a lowering of the basal metabolic rate and an improvement in the patient s condition It has been found that if the iodine is continued for a longer period the B M R rises again and the patient s condition deteriorates although the final state is often better than before the treatment was begun The use of iodine as a pre operative treatment has rendered thyroidectomy a comparatively safe operation and has resulted in enormous benefit to the sufferers from thyrotoxicosis

Goitrogenous Substances

Closely linked with the discovery of other antithyroid agents was the observation that certain substances when administered by mouth produce hyperplasia of the thyroid gland Thus Chesney et al (2) found that cabbage and other plants of the Brassica family when fed to rabbits cause thyroid hypertrophy The active agents appears to be nitrile derivatives and Marine et al (3) observed that acetonitrile when added to the food of rabbits produces enlargement of the thyroid gland

In 1941 the MacKenzies and McCollum (4) while engaged in a purely scientific enquiry chanced upon a discovery of considerable clinical importance They were investigating the possibility that sulphaguanidine when fed to rats on a purified diet containing synthetic B vitamins would prevent the synthesis of additional essential nutrients by the intestinal flora They observed that after periods varying from six to sixteen weeks a hypertrophy and hyperæmia of the thyroid gland with loss of colloid was invariably produced It was later found that this effect was not abolished by adding iodide to the diet but could be neutralised by adequate doses of thyroxine

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and then diiodotyrosine Sulphonamides effect their antithyroid action at this stage by competing with tyrosine for the iodine and so they inhibit the formation of diiodotyrosine Diiodotyrosine is next converted in the presence of a cytochrome enzyme into thyroxine This is stored in the thyroid as thyroglobulin a substance with a large molecular weight of 700 000 Thyroglobulin is acted on by a proteolytic enzyme and thyroxine with a molecular weight of 69 000 is now liberated Iodine is believed to produce its antithyroid effect partly by inactivating the proteolytic enzyme and partly by inactivating the T S H of the anterior pituitary

Clinical Applications All types of thyrotoxicosis respond to thiouracil treatment and as has been described by Himsworth (11) Nussey (12) Reveno (13) and others equally good results have been obtained in cases of primary and secondary Graves disease although the response may be slower with a toxic adenoma Grainger Gregson and Pemberton (14) on the other hand found that primary cases responded much more favourably than do secondary ones in fact the longer the duration of the goitre the less favourable the response to thiouracil All observers agree that patients who have recently received iodine show a delayed response when treated with thiouracil Thiouracil is equally efficacious in the treatment of thyrotoxicosis resulting from recurrence after thyroidectomy King and Rosellini (15) successfully treated a case of acute thyroiditis administering 200 mg of thiouracil three times daily The patient was free from symptoms in a week and was cured after a further two weeks treatment taking 200 mg daily Thiouracil has also been used by Raab (16) in angina pectoris and he found that 4 out of 10 cases were entirely free from symptoms during the treatment Thiouracil is considered by some to be more efficacious than iodine in the pre-operative treatment of thyrotoxicosis for by its use the surgeon can operate on a non toxic goitre Bartels (17) reports that it is especially of advantage in very severe cases of Graves disease especially those of long standing which either do not tolerate or fail to respond to iodine therapy Further by giving Lugol s solution after the B M R has been reduced to normal by thiouracil the operation may be rendered easier the gland being less vascular less hyperplastic and less friable Neither pregnancy nor diabetes are contraindications to the use of thiouracil In pregnancy it is wiser not

Kennedy and Purves (5) in the same year showed that rape seed and allyl thiourea were powerful goitrogenic substances and in 1943 the Mackenzies (6) found that thiourea and thiourea derivatives especially 2 thiouracil produced thyroid hyperplasia with inhibition of hormone production Astwood (7) used thiourea and thiouracil in the treatment of patients suffering from thyrotoxicosis and showed that the daily administration of 1 to 2 g of thiourea or of 0 2 to 1 g of thiouracil resulted in the relief of the patient s symptoms and a return to normal of the serum cholesterol and basal metabolic rate usually after a latent period of two to three weeks These results were rapidly confirmed by Williams and Bissell (8) and by Himsworth (9) Both thiourea and thiouracil have a similar action in cases of thyrotoxicosis but because of the action of side effects such as nausea vomiting and an unpleasant smell in the breath which may accompany the use of thiourea thiouracil methyl thiouracil or propyl thiouracil are the drugs now usually employed

Modes of Action Iodine sulphonamides and thiourea derivatives are believed to exert their antithyroid properties in different ways There is a balance between the activities of the pituitary and the thyroid for the thyrotropic hormone known also as the thyroid stimulating hormone or T S H liberated from the anterior lobe of the pituitary stimulates the thyroid to form thyroxine by increasing its power of taking up iodine and also by increasing the rate of the release of iodine probably as thyroxine This was shown by Stanley and Astwood (10) by means of radio iodine Thyroxine on the other hand inhibits the pituitary production of T S H The T S H in addition to increasing the output of thyroxine is thought to cause hyperplasia and increased vascularity of the thyroid gland and in some cases to accentuate the degree of exophthalmos

Radioiodine experiments also support the view that thyroxine is formed from tyrosine combined with iodine The synthesis is effected in certain stages Iodine is liberated from iodides in the presence of peroxidase manganese and oxygen Thiourea and its derivatives prevent the liberation of iodine from iodide by interfering with this peroxidase system and so block the formation of thyroxine Iodine thus liberated combines with tyrosine in the presence of a cytochrome enzyme to form first monoiodotyrosine

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which is usually in about four weeks the thiouracil dosage *is reduced to 100 mg three times or twice a day* When the B M R has fallen to between 0 and — 10 (Aub du Bois) a maintenance dose of between 100 to 200 mg daily should be given The dose must be sufficient to keep the B M R at this low level The progress chart of one of our patients a woman aged forty six years with a small hyperplastic goitre is given below —

THIOURACIL CASE SHEET

Date 194	B M R (Aub du Bois)	Blood Cholesterol mg/100 ml	Leucocytes per c.mm	Polymorphs	Pulse per min	Blood Pressure mm Hg	Wt lb	Thiouracil mg
Oct 25	+44	1 8	7 800	68	102	140/70	83	200 t i d
Nov 1	+ [illegible]	19	6 00	numerous	100	128/68	87	00 t i d
Nov 15	+ 0	190	8 700		78	13?/7?	96	200 t i d
Nov 21	+19	?18	9 00		84	110/80	99	200 t i d
Nov ?9	+14	46	6 300		88	11 /60	103	200 t i d
Dec 6	+ 7	236	8 800		84	120/65	108	100 b i d
Dec 13	+ 9	2 6	5 600		76	118/66	111	100 b i d
Dec 0	0	?50	10 000		76	125/62	117	100 b i d

A low white cell count before the commencement of thiouracil does not appear to be a contra indication to its use Thus in one of our cases the white cell count was 3 700 per c mm with 55 / polymorphonuclears before treatment and during treatment the count rose to 5 100 per c mm with 60 / polymorphonuclears This point has also been noted by Nussey (12) Auricular fibrillation may require simultaneous administration of quinidine although some cases are restored to normal rhythm by the use of thiouracil alone as recorded by McGavack et al (20) in 12 out of 18 cases If there is congestive failure it is wise to administer digitalis

Overdosage of thiouracil is shown by marked increase in the size of the thyroid gland or by symptoms of hypothyroidism such as a bloated appearance a feeling of heaviness lassitude and depression If there is any increase of exophthalmos during any stage of the treatment or if the goitre enlarges considerably gr 1 of dried thyroid should be given daily by mouth to diminish the output of thyrotropic hormone from the pituitary

Maintenance Treatment It is quite probable that the dosage of thiouracil will have to be frequently adjusted during the early

to continue the thiouracil during the last month but to substitute it then by iodine for in one case treated with thiouracil by Eaton (18) up to the confinement the child at birth had an enlarged thyroid Diabetes mellitus associated with thyrotoxicosis may be improved by thiouracil treatment less insulin being subsequently required but this is not so in every case for Rose and McConnell (19) treated a thirteen year old girl with thiouracil the result being that there was no increase in carbohydrate tolerance and no *diminution in insulin requirement*

Contra indications Thiouracil should be used with great caution if the goitre is causing sufficient pressure symptoms to *warrant surgical intervention* as a rapid enlargement of the gland might result in a catastrophe Thiouracil is contra indicated if the patient has to go abroad after the stabilising treatment is *completed where adequate supervision with blood counts and basal* metabolic readings are not available

Stabilising Treatment Some authorities do not keep their patients in bed during the initial period of treatment allowing them to continue at work providing adequate laboratory controls are carried out We prefer the patient to be in hospital during the stabilising treatment and in bed for the first three or four weeks Before the thiouracil treatment is begun the following observations should be made The weight of the patient the pulse rate the blood pressure the circumference of the neck the basal metabolic rate the total white cell count and percentage of polymorphonuclear *cells and the blood cholesterol The drug* is given in tablet form by mouth 200 mg b i d In some cases a satisfactory response is not obtained until the dose is raised to *200 mg t i d* The white *count is repeated twice a week* during the early stages of treatment the other observations apart from the daily temperatures and pulse record being carried out weekly Dried yeast tablets (D C L) two t i d may be given during the treatment with the idea of preventing the development of agranulocytosis but they are of *doubtful* value *The first effects of the* thiouracil treatment are usually seen within one or two weeks the lag being due to the time taken to use up the stores of thyroid hormone and prevent *the formation* of more In some cases no improvement is noted for four to twelve weeks especially if iodine has been recently given As soon as the B M R is falling satisfactorily

by which is meant a white cell count below 3 000 per c cm without evidence of infection Penicillin should be given in any case of leucopenia to prevent the secondary infection which so often proves fatal Thus Rothendler and Vorhaus (24) successfully treated a case by injections of 20 000 units of penicillin every three hours for four days together with daily injections of 2 ml of crude liver and a whole blood transfusion of 500 ml

Other toxic reactions make a formidable list but fortunately are not often seen They include drug fever usually about the eighth day morbilliform or urticarial rashes allergic arthritis nausea vomiting abdominal pains diarrhœa headache enlargement of lymph glands or of the submaxillary glands œdema of the legs jaundice of an obstructive type increase of exophthalmos thrombocytopenic purpura acute hæmolytic anæmia heart block mental stupor confusion and delusional insanity

METHYL THIOURACIL AND PROPYL THIOURACIL

These substances were introduced as being less toxic and more potent than thiouracil Radioiodine and clinical tests indicate that methyl thiouracil is from two to one and a half times as effective as thiouracil whereas propyl thiouracil is probably no more effective than thiouracil dose for dose The initial dose for methyl thiouracil is usually 100 mg b i d and for propyl thiouracil 100 mg t i d The dosage is subsequently reduced when the B M R has fallen to maintain it at a figure between 0 and — 10 Himsworth (25) considers there is little to choose between the toxicity of these three drugs The American workers such as Beierwaltes and Sturgis (26) prefer propyl thiouracil but methyl thiouracil is often employed in England and is the preparation we use Propyl thiouracil is certainly not free from toxic effects Agranulocytosis developed in the first two cases in which we used it in doses of 50 mg t i d but neither proved fatal Bartels (27) in America also reported three cases of agranulocytosis and eleven of leucopenia during propyl thiouracil treatment Juliar and Harris (28) have further described a fatal case of agranulocytosis due to propyl thiouracil

The Combined Use of Iodine and Thiouracil Derivatives Whether iodine should be simultaneously administered with a thiouracil derivative in the routine treatment of thyrotoxicosis is a

stages of the maintenance treatment The B M R and leucocyte count should be determined every month until the condition is firmly stabilised subsequently three monthly readings should suffice As a routine measure every case should be treated continuously for ten months

Toxic Reactions These have been met with in from 10 to 20% of cases treated *but with the general use of smaller doses than were originally prescribed they appear to be less frequent* The most dangerous complication is agranulocytosis and unfortunately it may appear without any warning and rapidly prove fatal despite regular control of the dosage of thiouracil by B M R and white cell readings In a survey of 1 091 cases treated at eleven clinics in America and at University College Hospital London Moore (21) reported that 10 patients developed agranulocytosis of which 3 proved fatal It is therefore probable that the mortality from agranulocytosis is about ¼ % or less and its *importance must not be over exaggerated* *Wosika and Braun (22) reported a fatal case in a woman aged fifty years who had taken* a total amount of 29 1 g of thiouracil during eleven weeks in average sized doses The response of the thyrotoxicosis to the thiouracil had been good The symptoms of the agranulocytosis consisted of fever sore throat with redness and a dirty white exudate and restlessness The patient died in coma in six days the white cells falling to 500 per c mm with a complete absence of neutrophils The usually accepted forms of treatment were of *no avail pentnucleotide yellow bone marrow ascorbic acid thiamine hydrochloride whole blood liver folic acid and penicillin* Cargill and Lesses (23) had a fatal case just a year after the initiation of treatment which had been most carefully controlled throughout The development of agranulocytosis appears to be a matter of idiosyncrasy unpredictable and probably unpreventable but fortunately rare The patient should always be warned to discontinue the thiouracil directly he feels a sore throat enlarged cervical glands or malaise *or if he develops a cold or skin rash He should then immediately report to his doctor for a blood count Moore (21) found that there is a period of maximal danger of agranu*locytosis between the fourth and eighth weeks of treatment so that at this stage particular vigilance is necessary and it is also at this period that there is the greatest likelihood of leucopenia

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matter of opinion We do not do so Beierwaltes and Sturgis (26) recommend that 5 minims of Lugol s solution should be given daily together with propyl thiouracil They claim that if an operation is subsequently performed the thyroid is less friable and bleeds less easily and further as the iodine acts at a different point in the formation of thyroxine than does thiouracil the restoration to health of the patient is accelerated Astwood (29) however believes that iodine should not be used with a thiouracil derivative in the treatment of thyrotoxicosis as there is a high frequency of recrudescence of thyrotoxicosis during prolonged treatment with the two substances

Pre Operative Treatment There is here also a difference of opinion Should iodine be used alone or combined with a thiouracil derivative ? Beierwaltes and Sturgis (26) recommend that 100 mg of propyl thiouracil should be given t i d a c together with 4 drops of iodine solution daily until the B M R has fallen to zero The operation should be performed provided also that the clinical features of thyrotoxicosis have been controlled No antithyroid drugs are required after the operation This treatment is suitable both for cases of exophthalmic goitre and of toxic adenoma Bartels (30) says that the pre operative use of propyl thiouracil has reduced the operative mortality in surgical thyroid clinics from 3% to 0 1 % and it should always be used in severe cases We are accustomed to prepare our patients for operation by the use of iodine alone except in very severe cases in which a satisfactory response cannot be obtained In such instances we believe that methyl thiouracil should first be used and if a satisfactory response results iodine is also prescribed during the last two weeks of the pre operative treatment

Results of Treatment with Thiouracil Derivatives A satisfactory assessment of the results of thiouracil treatment can only be made after the patient has discontinued the drug for several years In the majority of the reports the figures are based on periods of one or two years observation or less and there is considerable variation in the results observed Danowski et al (31) used thiourea and iodine as they consider that thiourea is not only at least as effective as thiouracil but also less toxic One hundred and eighteen patients were so treated and of these 89 were followed for periods of six months to more than two years Eight patients

were observed for five to sixteen months following a course of thiourea which had lasted from 6 to 24 months The B M R remained satisfactory in all of these cases but there was recurrence of clinical symptoms of hyperthyroidism in one of them Poate (32) believes that methyl thiouracil is superior to other thiouracil preparations and in a series of 200 cases considered there is an 80 % chance of cure without operation The average period of treatment was nine months and with a large thyroid a year or longer might be required If thyroideum siccum has also been used the maintenance treatment should be prolonged for another two to four months Verel (33) observed a series of 62 patients treated with thiouracil or methyl thiouracil for periods between six months and four and a half years In all cases the thyrotoxicosis was successfully controlled and the majority were back in full employment within six weeks of starting treatment Thyroidectomy was performed in two cases in one instance on account of persistent granulopenia in the other because of excessive thyroid enlargement In 33 cases the result was satisfactory after the thiouracil had been discontinued for three months or longer One patient developed myxœdema after being off thiouracil for nineteen months In 5 cases in which no thiouracil was taken for thirty three to thirty nine months no relapse occurred Ravdin et al (34) believe that sustained remissions after treatment for six to twelve months can be expected in about 50 % of cases McCullagh and Sirridge (35) found recurrence of hyperthyroidism in 2 cases out of 6 who had been treated with methyl thiouracil and iodine for periods of five to twelve months with complete control Meulengracht and Kjerulf Jensen (36) treated 190 patients with methyl thiouracil for an average period of twelve months An examination was subsequently made of 111 patients who had been without treatment for three months and in 10 cases relapse had occurred Unfavourable results have also been reported by Starr et al (37) who treated 28 patients with propyl thiouracil for periods up to ten months and in nearly every case relapse occurred on discontinuing the drug Frisk (38) in a series of 126 patients treated with methyl thiouracil found that 14 % had a remission of at least eighteen months duration

It is quite clear from the above figures that the tendency to relapse is considerable but when prolonged remissions do occur

several years must pass after discontinuance of treatment before the real efficiency of the thiouracil drugs can be determined. When however relapse does occur the symptoms of thyrotoxicosis are usually amenable to a further course of treatment.

RADIOACTIVE IODINE

Radioactive iodine with a mass of 131 and a half life of eight days is used both for the diagnosis and treatment of thyrotoxicosis. It is said to have a half life of eight days because at the end of that period it has lost 50% of its activity. Taken by mouth as a colourless solution in water it is quickly absorbed from the stomach. The greater part of the absorbed radioiodine is concentrated in the thyroid. It is essential that the patient shall have taken no iodine for at least four weeks before the tests for thyrotoxicosis are made.

Diagnosis of Thyrotoxicosis. The percentage uptake of radioiodine is determined by giving by mouth a tracer dose of 30–50 microcuries of I^{131}, the percentage amount present in the thyroid being estimated by a Geiger Muller counter at twenty four hours. Normally the gland retains 30 to 40%, in hyperthyroidism between 40% to 90% is present in the thyroid and in hypothyroidism the figure is between [illegible] and 10%. There is some overlap of these values for at times in thyrotoxicosis less than 30% is retained in the thyroid at twenty four hours. In one of our cases of diffuse thyrotoxicosis the B.M.R. was + 17 (Aub du Bois) and the percentage iodine uptake was 90. *The diagnosis of thyrotoxicosis may also be made by the plasma clearance test.* A similar tracer dose of I^{131} is given by mouth and the rate of clearance of the radioiodine from the *blood is determined.* Normally about 5–40 ml of plasma are cleared of iodine per minute; in thyrotoxicosis the clearance rate is raised to about 100–1400 ml per minute and in hypothyroidism the figure is low, usually less than 5 ml per minute.

The Treatment of Thyrotoxicosis. A tracer dose of 30–50 microcuries of I^{131} is given by mouth and the percentage uptake at twenty four hours is determined. From this figure and the estimated weight of the thyroid the therapeutic dose is calculated. This is usually equivalent to 100–200 microcuries per gram of thyroid, the total dose being between 1–5 or 10 millicuries. The normal weight of the thyroid is about *20* grams. The beta particles of radioiodine exert *their effect locally in the thyroid* with a range of

about 1-4 mm The gamma rays carry a distance of about one metre and are picked up by the Geiger Muller counter

Special precautions must be taken when radioiodine is administered Space does not allow detailed rules to be given here but they are usually drawn up by the special departments concerned with the investigation and treatment of patients with radioactive substances The patient should be in a single room Contamination with the drug is most likely to occur through the urine or if the patient is sick through the vomit Bed clothing night clothes handkerchiefs bed pans lavatories or baths may be affected Nurses and attendants must wear gloves gowns and masks when attending to the patient dealing with bed pans or cleaning the room Crockery must be kept separate The urine is not thrown away until the radioactivity is considerably diminished

Various reports on the results of treatment of patients suffering from thyrotoxicosis have been published by Hertz and Roberts (39) in 1946 by Prinzmetal et al (40) in 1949 and by Gordon and Albright (41) in 1950 Some authorities consider that the diffuse thyrotoxic goitre is more suitable for radioiodine treatment than is the toxic adenoma Gordon and Albright (41) treated 83 cases of diffuse goitre and 37 of nodular goitre and found the response in both types similar Owing to the error in estimating the size of the thyroid it appears safer to give a small dose of 3 millicuries and after two to three months to repeat the dose if the thyrotoxicosis is not arrested Gordon and Albright (41) found that 59 of their patients responded satisfactorily to a single dose of this size 36 patients required 2 doses 16 patients were given 3 doses and 7 patients needed 4 doses Only 2 patients had more than 4 doses

Radioactive iodine is of value not only in the treatment of primary and secondary Graves disease but also in thyrotoxicosis associated with severe heart failure in cases complicated by extreme emotional disturbances and in multiple recurrences after what appeared to be adequate surgical removal It should not be used during pregnancy after the fourth month or when the goitre is large hard or nodular Chapman and Evans (42) found that the degree of exophthalmos changes very little after radioiodine treatment but the appearance of the eyes is improved from loss of chemosis lid retraction and œdema

Complications It is too early to know whether carcinoma of

the thyroid may develop after many years in the gland which has been treated with radioiodine There appears to be no danger of causing sterility In the early days of treatment radioiodine was limited to patients over the age of forty five years now it is given to patients of any age Myxœdema may result from overdosage but this can be controlled by the administration of thyroideum siccum No damage to the kidneys has been noted

Treatment of Carcinoma of the Thyroid Radioiodine is only taken up by certain types of carcinoma of the thyroid In all cases to be treated as complete a surgical removal of the gland as possible should be made first and then sufficient radioiodine given to induce a complete state of myxœdema which is subsequently controlled by the oral administration of thyroideum siccum Good results may be obtained in the treatment of secondary bony deposits of carcinoma of the thyroid A preliminary thyroidectomy or the *administration of thyrotropic hormone appears to assist the uptake* of radioiodine by the secondary deposits

Conclusions It is not claimed that thiouracil or its derivatives remove the cause of thyrotoxicosis and there is little difference between the risks of medical and surgical treatment in Graves disease The operative mortality rate in the best hands is below 1% and the chance of death from the thiouracil derivatives is possibly a little less It is said that in the exophthalmic class of Graves disease the chance of cure is as high as 80% whereas with thyroidectomy the curative rate is higher up to 85% The likelihood of a permanent cure with thiouracil has been discussed above The chief drawbacks of thyroidectomy are the possible complications of the operation especially tetany recurrent laryngeal nerve palsy and the subsequently development of exophthalmic ophthalmoplegia Further relapse may take place as long as twenty years after the operation Thyroidectomy is especially indicated *in toxic adenoma and the high percentage of carcinomas* which develop in fœtal adenomas of the thyroid is another point in favour of operation Medical treatment with thiouracil derivatives *is indicated* in the elderly and in patients unsuitable for surgery owing to cardio vascular *degeneration* or who have had a previous thyroid operation Thiouracil derivatives are also useful as a pre operative treatment in those who have become iodine fast owing to prolonged iodine treatment They can be recommended in cases

in which the goitre is not large or in those who refuse surgery An operation should be advised if during thiouracil treatment a large goitre persists despite the administration of thyroideum siccum Finally the cosmetic effect resulting from the removal of a large goitre cannot be achieved by thiouracil or its derivatives the maintenance treatment is apt to be difficult with thiouracil and there remains the never ending possibility of the sudden development of agranulocytosis

It is too early to assess the ultimate results of treatment with radioiodine but it may become the method of choice in the majority of cases Elaborate equipment and a specially trained team of workers are required and so it is not suitable for use in the general wards of a hospital or in private practice

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CHAPTER XI

THE NERVOUS SYSTEM

Electro encephalography

It has been known for over sixty years that electrical currents are produced in the brains of animals both at rest and during stimulation In 1891 Gotch and Horsley (1) recorded such currents produced in the exposed brains of animals using a string galvano meter or a capillary electrometer Berger (2) in 1929 first showed that changes in electrical potential arising in the cerebral cortex could be demonstrated in man with electrodes applied to the scalp He used originally a Siemen s galvanometer and the deflections were very small There was at first some doubt as to whether the electrical changes did actually arise in the brain but in 1934 Adrian and Matthews (3) confirmed Berger s findings and demon strated that the waves recorded arose in the parieto occipital region in both sides of the brain near Vogt s area 19 all extraneous causes of change in potential such as muscular activity eye move ments and electrode artefacts were excluded Electro encephalo graphy is comparable with electro cardiography in technique but the potential changes led off from the scalp are often more than a hundred times smaller than those recorded from the body surface in electro cardiography Thus the normal scalp potential changes are in the region of 5 to 100 microvolts but occasionally under abnormal conditions they may rise to 1 millivolt (1 microvolt = 1 millionth of a volt) The electrical potential changes on the brain surface are ten to fifty times greater than those obtained on the scalp

The Electro encephalograph There are many different types of electrode the most usual ones being either a saline pad which may be moved about the head at will or a solder pledget sealed to the scalp with collodion The second is commonly used for prolonged investigation The different techniques used have been fully described by Gibbs Davis and Lennox (4) Kornmuller (5) Walter (6) Balado Romero and Noiseux (7) and others

The hair should be cut short or pushed aside under the electrodes It should be moistened if it is very dry and the portion of

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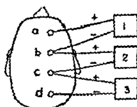


Fig 49 Paired electrodes (after Williams and Gibbs (8))

scalp under the electrodes should be cleaned with alcohol Either paired electrodes or a single scalp electrode system may be employed In the paired electrode system four electrodes (a b c and d) are placed on the scalp connected with three amplifiers (1 2 and 3) as shown in the diagram (see Fig 49) The amplifiers are thermionic valves whereby an amplification of more than a millionfold can be obtained The potentials thus amplified are led to three recording oscillographs

Recording can also be carried out on paper by an ink writing oscillograph or photographically by means of cathode ray tubes As the frequencies encountered are relatively slow the ink writing oscillograph in virtue of its simplicity and economy of time and money is superseding the more unstable cathode ray tube

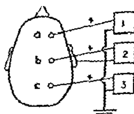


Fig 50 Single electrode (after Williams and Gibbs (8))

Single scalp electrodes are employed with an indifferent electrode placed on the ear and earthed The arrangement of a three single electrode system is shown in the diagram (see *Fig 50*) a b and c are electrodes placed on the scalp and 1 2 and 3 are amplifiers

The single scalp electrode records the potential change occurring at a single point in the brain and gives a truer reproduction of a pure wave than do the paired electrodes It therefore records accurately the abnormal wave forms which are encountered in grand mal petit mal and psychomotor epilepsy The paired electrodes are used to localise foci of activity for with paired electrodes changes in potential common to both electrodes are eliminated and although a true picture of the electrical events occurring in the cortex is not given the local discharges are rendered more prominent As Williams and Gibbs (8) point out if the potential gradient is visualised as a hill the single scalp

electrode records the height of the hill above ground level represented by the indifferent electrode whereas the paired electrodes measure the difference between the two points on the hill under examination without indicating the actual height of either point above the ground level Thus with the single scalp electrode system when the hill top or greatest change in potential is placed under one electrode whichever way the elec trode is moved the potential will fall off as shown by the recorded

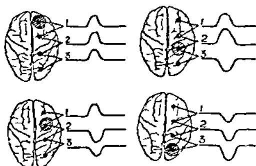


FIG 51 Localisation of focus of activity by paired electrodes (after Williams and Gibbs (8))

deflections With paired electrodes if the hill top is at the end of the row of electrodes the potential will fall in the same direction in each pair and will cause a deflection of each of the recorders in the same direction the maximum deflection occurring from the electrode situated over the focus of activity If how ever the hill top occurs at one of the middle electrodes the fall will be away from that electrode in both directions and so will pass through the circuit on each side in opposite directions making an upward signal or deflection in one and a downward in the other the waves are then said to be out of phase indicating that the focus of activity is situated under or adjacent to the middle electrode common to the two pairs

The application of this method for the localisation of the focus of activity by means of paired electrodes is shown in the diagram (see Fig 51)

The Alpha Waves or Berger Rhythm Berger placing one electrode on the frontal and the other on the occipital region

demonstrated the occurrence of waves arising from the brain in health. They have an amplitude of 10 to 100 microvolts and a frequency of about 10 Herz (10 cycles per second). In childhood the frequency is about 6 per second and after the age of fifty the frequency diminishes. They are present when the eyes are shut and usually disappear when the eyes are open. In a few people they are not present. They are nearly sinusoidal and come and go at regular intervals (see Fig. 52A). When the eyes are shut mental concentration or a sudden noise will cause the wave to diminish in amplitude. Rarely, with sufficient mental relaxation, the waves persist even with the eyes open. The alpha waves are replaced by other wave forms during sleep. They are present in blind people, but noises or tactile stimuli may cause their disappearance. The persistence of the waves therefore appears to depend upon that part of the cerebral cortex which is concerned with the integration of the visual stimuli. They are thought to be produced by groups of neurones acting in unison, their synchronisation being due to a local pace maker whose activity is modified when the neurones are brought into action. Thus they are evident when cortical activity is suppressed and abolished during functional activity. Lemere (9) has used this method to distinguish between blindness and hysterical blindness or malingering. In true blindness the alpha waves persist when the eyes are opened and the person attempts to look at an object; in feigned or hysterical blindness the alpha waves cease on opening the eyes and looking at an object. This suggests that in hysteria neither the sensory nor motor pathways nor the cortical centres are affected.

The Beta Rhythm. Berger also described beta waves occurring in health. They are smaller than the alpha waves and have a frequency of 18 to 25 per second. They are believed to arise in the pre-frontal region of the cerebral cortex. Similar waves can be produced by contraction of the frontal and temporal muscles and they are more prominent in records from individuals with wrinkled foreheads and raised eyebrows. Little is known about these minute waves and their significance is not yet understood.

Changes in the Electro encephalogram in Health

Sleep. Loomis, Harvey and Hobart (10) have shown that the alpha waves persist during the preliminary drowsy stage while the

subject is still conscious With the onset of sleep the alpha waves cease and intermittent groups of waves appear known as spindles with a frequency of 14 or 15 per second Later in addition to the spindles' larger and more prolonged waves known as delta waves are seen A dream may be accompanied by the appearance of alpha waves

Hypnosis The alpha rhythm is sometimes slowed during hypnosis but the electro encephalogram does not resemble that of natural sleep

Cerebral narcotics cause the appearance of slower and larger waves and basal narcotics result in quicker and larger waves

The Electro encephalogram in Disease

Studies have been chiefly directed to changes seen in the electro encephalogram in increased intracranial pressure in cerebral tumours and in the epilepsies

Increased Intracranial Pressure This may be due to obstruction of the ventricular system to cerebral tumours concussion or meningitis Berger (11) in 1931 showed that slow delta waves can be detected all over the skull The waves may have an amplitude up to 100 microvolts and a frequency of 3 per second Williams (12) has shown that the abnormal waves are not directly related to the raised intracranial pressure but are caused by changes in the water content of the cerebral hemispheres which is probably limited to the white matter If the increased pressure is due to a cerebral tumour reduction of the pressure by intravenous injection of hypertonic sodium chloride solution may enable the site of the tumour to be located as the slow waves are now confined to the site of the tumour as shown by Walter (6)

Cerebral Tumours In 1933 Berger (13) demonstrated similar slow waves arising over the affected part of the brain in two cases of cerebral tumour The waves appeared when one of the electrodes was placed over the tumour and they were not present when the tumour was midway between the electrodes In 1935 Kornmuller (5) and Foerster and Altenburger (14) demonstrated that the potentials arise in the surrounding cerebral tissue and not in the tumour itself Walter (15) has evolved a method for locating certain tumours by means of the electro encephalogram

Walter s method of localising a cerebral tumour with paired

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lesion was demonstrated by operation or by autopsy and in 15 the diagnosis was established by other means A single focus was found in 36 cases and all were correctly located In 7 cases two foci were found in 4 of which the pathological cortical areas were confirmed In 7 cases generalised abnormal waves were found In 41 cases of suspected intracranial lesions the electro-encephalogram was normal and further clinical investigations revealed no abnormality

In general it may be stated that any tumour causing progressive destruction of cortical tissue will cause abnormally large slow waves of electrical potential to arise in the damaged cortex Similarly any other lesion such as an abscess or a demyelinating disease resulting in cerebral dysfunction will be associated with identical wave forms It follows that discrete and non progressive cortical lesions or deep tumours not affecting the cerebral hemispheres or the cortex itself or cerebellar tumours are unlikely to be unmasked by indirect electro-encephalography Walter and Dovey (16) have described a method using a needle electrode by which the electrical activity of the subcortical structures can be determined This can be used to supplement conventional methods of identifying and locating subcortical tumours at operation

Epilepsy In 1937 Golla Graham and Walter (17) showed that there are changes in the electro-encephalogram in 50% of patients under the age of forty suffering from convulsive epilepsy even though no fit is seen at the time of examination The majority of such abnormal electro encephalograms occurred in patients suffering from grand mal An abnormal electro encephalogram is rarely seen in a patient who first has epileptic fits after the age of forty The abnormality between fits consists in the occurrence of intermittent irregular slow waves Walter states that in grand mal the focus of discharge is usually found in the region of the superior frontal gyrus on one or both sides Krynauw (18) says that foci of discharge are also quite commonly post central or even occipital in grand mal and other workers have been unable to find a predilection for any particular cortical area In petit mal the abnormal waves are widespread but the focus is usually post central Seizure waves occur during a fit the focal discharges becoming greater and the area of discharge spreading

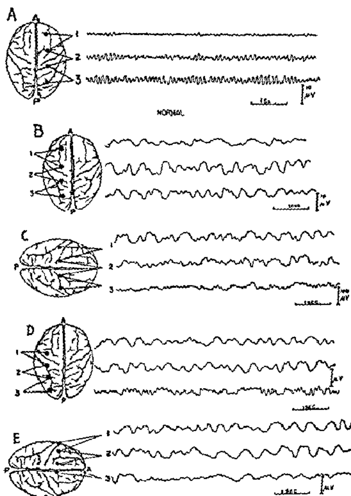


FIG 52 Method of localisation of a cerebral tumour with paired electrodes (after Williams and Gibbs (8))

electrodes is illustrated in the diagrams (see Fig 52) reproduced from an article by Williams and Gibbs (8) There are five stages in the investigation A Right half of brain normal B Abnormal focus in left parietal region C Abnormal focus confirmed in left side of brain D Abnormal focus localised antero posteriorly E Abnormal focus localised transversely

Williams and Gibbs (8) investigated 50 cases in 35 of which the

The types of waves met with in epilepsy are illustrated in the accompanying diagram (see Fig 53)

Head Injuries Williams (23) has investigated with the electro-encephalograph 74 patients suffering from acute head injuries due to war wounds The tests were made within twenty days of the injury He found that widespread abnormally slow waves suppression of normal frequencies and outbursts of high voltage

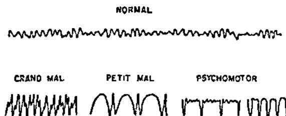


Fig 53 Types of waves in epilepsy (Lennox (18))

sine waves with a frequency of 2 or 3 per second indistinguishable from sub clinical epileptic attacks occurred in the acute stage Further the degree of abnormality corresponded with the clinical state of the patient and progressive changes indicated improvement or deterioration The epileptic type of waves bore no prognostic significance as regards the occurrence of post traumatic epilepsy Williams (23) also investigated the electro encephalograms in chronic post traumatic states following head injuries The electro encephalogram was abnormal in 50% of cases and there was a definite relation between the electro encephalogram the severity of the injury the persistence of symptoms and the presence of dural penetration

Psychopathic Disorders Attacks of uncontrollable anger or negativism and behaviour disorders in children may be accompanied by cerebral dysrhythmia and an abnormal electro encephalogram The electro encephalogram will also indicate whether the cerebral cortex is affected in cases of Sydenham s chorea

Conclusions Electro-encephalography which started life as suspect is now accepted as a genuine method of studying electrical changes in the cerebral cortex It has already become a useful adjunct to other clinical methods in investigating the site of cerebral tumours and abscesses and it has special value in giving

The waves are then slower but larger and more irregular Transitory bursts of seizure waves are often seen in the absence of a fit and Lennox (19) has called them subclinical seizures An increase in the number of these outbursts often precedes a grand mal fit by days and can be used in predicting the onset of a convulsion Gibbs Gibbs and Lennox (20) describe the seizure waves in grand mal as sharp spikes occurring at twenty five to thirty per second in petit mal there are quick sharp spikes alternating with slow round waves and in psycho motor attacks (psychic variants) square flat waves occur at about three to four per second As epilepsy is associated with the development of abnormal rhythms in the cerebral cortex they have called it a paroxysmal cerebral dysrhythmia According to Gibbs (21) about 10 % of normal people show a cerebral dysrhythmia indistinguishable from that occurring between the seizures in epilepsy and it is possible that such individuals are suffering from asymptomatic epilepsy Further about 5 % of the near relatives of epileptics show an abnormal electro encephalogram Despite this corroborative evidence of petit mal provided by an electro encephalogram was successfully used by the defence in a murder case in England in 1942 when prolonged clinical observation of the accused in prison showed no evidence in epilepsy

Symptomatic Epilepsy There is no difference between the record of a fit resulting from trauma tumour or any other lesion and that seen in idiopathic epilepsy The fit may not necessarily be associated with any focal electrical abnormality

Hysterical Fits No abnormality is seen in the electro encephalogram during the fits and except that unstable people are more liable to show abnormalities in the electro encephalogram than are stable subjects the resting record is usually perfectly normal

Golla McKissock and Walter (22) have shown that surgical removal of the portion of the cerebral cortex in which the delta waves originate may abolish the fits for a few months but they ultimately recur The actual origin of the fit therefore appears to be due to generalised cerebral changes

Anti convulsant drugs such as bromides and Luminal which are used for the treatment of epilepsy reduce the number of abnormal electrical outbursts or subclinical fits especially in grand mal epilepsy

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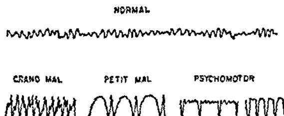


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objective proof of the presence of epilepsy and the exact type of fit in subjects whose history is inconclusive. It is of value in demonstrating impairment of function of the central nervous system in cases of head injuries. It is also helpful in the diagnosis of narcolepsy, subdural hæmatoma, cerebral trauma, meningitis, encephalitis, Schilder's disease and disorders of behaviour. Severe degrees of dementia may occur with a normal electro encephalogram. For a more detailed discussion of electro-encephalography the reader is referred to the book edited by Hill and Parr (24).

Thymectomy in Myasthenia Gravis

In 1901 Weigert (25) recorded a case of myasthenia gravis with hyperplasia of the thymus. Since then numerous observations have been made and it appears that the thymus is always persistent, hypertrophied, or the seat of a benign tumour in myasthenia gravis. Norris (26) suggests that in myasthenia gravis the thymus shows specific histological changes, consisting of hyperplasia of the epithelial cells of the medulla. In the hyperplastic areas the normal structure of the gland may be preserved, or an undifferentiated syncytial mass of epithelium is found obliterating the round cells and Hassall's corpuscles, its structure closely resembling that of the fœtal thymus. The causal connection which is now well established between changes in the thymus and myasthenia gravis is unknown, but it has been suggested that a substance is formed in the thymus which inhibits the cholinergic transmission from nerve to muscle. Sauerbruch (27) in 1912 introduced the operation of thymectomy for myasthenia gravis; the patient was also suffering from hyperthyroidism. Blalock et al. (28) in 1941 reported on thymectomy in 6 cases of myasthenia gravis. One patient died, 3 were symptom free three months after operation, one was improved but still required prostigmin, and one was unchanged two months later. Viets (29) reports on 36 patients suffering from myasthenia gravis treated by thymectomy at Massachusetts from 1941 to 1944. Seven of these patients had thymomas and 29 showed various degrees of involution and the formation of germinal centres in the thymic tissue. Good to excellent results were obtained in about 50% of the cases. Neostigmine may be given with advantage intravenously during the operation, 0.5 mg. of neostigmine methylsulphate intravenously being

approximately equivalent to 15 mg of neostigmine bromide by mouth

Keynes (30) has done much to lower the operative mortality of thymectomy which should now be below 5 / He obtained the best results in cases with a short history Of 1?0 patients who did not have thymic tumours 79 have shown a complete or almost complete remission of symptoms Keynes is opposed to a transpleural operation as even temporary collapse of the lung is harmful to the myasthenic patient He also emphasises the danger of giving an enema to a patient after thymectomy almost invariably the patient will collapse with syncope and such an experiment should never again be made Fatalities after operation are nearly always due to pulmonary complications A myasthenic crisis may cause excessive bronchial secretion with pulmonary consolidation and death Viets however has not seen such a myasthenic crisis and considers that when patients drown in their own secretion it is due to excessive or ill considered use of neostigmine Primary operation in the case of thymomas was found to be very unsuccessful The operation here is more difficult and although the immediate result may be good a serious and irrevocable relapse is very liable to occur Such cases are best treated primarily by deep X rays Later it may be possible to remove the thymus with the tumour

Conclusion It is probable that thymectomy is indicated in rather less than 50 / of all cases of myasthenia gravis It is contra indicated in cases well controlled by neostigmine or by tetra ethylpyrophosphate With neostigmine remissions may be observed equal in length to those following thymectomy Further tolerance to neostigmine is unlikely to occur

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(3) Adrian and Matthews *Brain* 1934 57 355 and *Journ of Physiol* 1934 81 440

(4) Gibbs Davis and Lennox *Arch Neurol and Psych* 193 34 1133

(5) Kornmuller *B ol Rev Camb Philos Soc* 1935 10 383

(6) Walter *Proc Roy Soc Med.* 193? 30 579 and *Journ Neu ol and Psych* 1938 N.S I 3 9

(7) Balado Romero and Noiseux *El Electro encephalograma humano* 1939 Buenos A res

(8) Williams and Gibbs *Arch Neurol and Psych* 1939 41 519

(9) Lemere *Journ Amer Med Assocn* 194? 118 884

objective proof of the presence of epilepsy and the exact type of fit in subjects whose history is inconclusive It is of value in demonstrating impairment of function of the central nervous system in cases of head injuries It is also helpful in the diagnosis of narcolepsy subdural hæmatoma cerebral trauma meningitis encephalitis Schilder s disease and disorders of behaviour Severe degrees of dementia may occur with a normal electro encephalogram For a more detailed discussion of electro encephalography the reader is referred to the book edited by Hill and Parr (24)

Thymectomy in Myasthenia Gravis

In 1901 Weigert (25) recorded a case of myasthenia gravis with *hyperplasia of the thymus Since then numerous observations* have been made and it appears that the thymus is always per *sistent* hypertrophied or the seat of a benign tumour in myas thenia gravis Norris (26) suggests that in myasthenia gravis the thymus shows specific histological changes consisting of hyper plasia of the epithelial cells of the medulla In the hyperplastic areas the normal structure of the gland may be preserved or an undifferentiated syncytial mass of epithelium is found obliterating the round cells and Hassall s corpuscles its structure closely resembling that of the fœtal thymus The causal connection which is now well established between changes in the thymus and myas thenia gravis is unknown but it has been suggested that a substance is formed in the thymus which inhibits the cholinergic transmission from nerve to muscle Sauerbruch (27) in 1912 introduced the operation of thymectomy for myasthenia gravis the patient was also suffering from hyperthyroidism Blalock et al (28) in 1941 reported on thymectomy in 6 cases of myasthenia gravis One patient died 3 were symptom free three months after operation one was improved but still required prostigmin and one was un changed two months later Viets (29) reports on 36 patients suffering from myasthenia gravis treated by thymectomy at Massachusetts from 1941 to 1949 Seven of these patients had thymomas and 29 showed various degrees of involution and the formation of germinal centres in the thymic tissue Good to ex cellent results were obtained in about 50 % of the cases Neostig mine may be given with advantage intravenously during the opera tion 0 5 mg of neostigmine methylsulphate intravenously *being*

CHAPTER XII

THE HÆMOPOIETIC SYSTEM

Types of Anæmia and their Treatment

As our knowledge of the causation of the various anæmias increases ætiological classifications become more satisfactory The following classification is based on that of Davidson (1)

1 *Nutritional Deficiency Anæmias* These may be due to (*a*) Lack of the anti anæmic or hæmopoietic principle This is a primary defect (the diet being adequate) in pernicious anæmia and a secondary defect (*i e* resulting from a recognisable cause) in the anæmias of sprue and of dysentery in tropical megalocytic anæmia in the pernicious anæmia of pregnancy in Diphyllo bothrium latum infestation in carcinoma of the stomach and after gastrectomy In the latter group there is also often an error in iron assimilation There may be a deficiency of storage of the hæmopoietic factor in advanced cirrhosis of the liver (*b*) Lack of the hæmopoietic factors such as iron copper thyroxine vitamin C and a proper salt balance in the food This deficiency may be primary as in simple achlorhydric anæmia and the Plummer Vinson syndrome in which the food intake is often satisfactory but the absence of hydrochloric acid results in impaired absorption of iron or secondary to a recognisable food or endocrine defect as in the simple nutritional anæmia of infants chlorosis chronic gastritis and enteritis cœliac disease starvation prolonged milk feeding as in the treatment of peptic ulcers and in myxœdema

2 *Post hæmorrhagic Anæmias* These occur after acute or chronic hæmorrhage

3 *Hæmolytic Anæmias* (*a*) Acute hæmolysis may result from blackwater fever malaria paroxysmal hæmogolobinuria septi cæmia or toxins such as snake venoms It also occurs in Lederer s anæmia (*b*) A persistent hæmolysis as in congenital or acquired acholuric jaundice sickle cell anæmia Cooley s anæmia Von Jaksch s anæmia and lead poisoning

4 *Anæmias due to Inhibition of the Bone Marrow Function* These may be primary as in aplastic anæmia or secondary as in aplastic anæmia due to agents such as X rays radium benzol

(10) LOOMIS HARVEY and HOBART *Science* 1937 86 448
(11) BERGER *Arch f Psych* 1931 94 16
(12) WILLIAMS *Brain* 1939 62 321
(13) BERGER *Arch of Psych* 1933 100 301
(14) FOERSTER and ALTENBURGER *Deutsch Ztschr f Nervenh* 1935 135 277
(15) WALTER *Lancet* 1936 ii 305
(16) WALTER and DOVEY *Lancet* 1946 i 5
(17) GOLLA GRAHAM and WALTER *Journ Ment Sci* 1937 83 137
(18) KREYNAUW *Brit Med Journ* 1939 ii 100
(19) LENNOX *Journ Amer Med Assocn* 1939 114 1347
(20) GIBBS GIBBS and LENNOX *Brain* 1937 60 377
(21) GIBBS *Journ Amer Med Assocn* 1942 118 216
(22) GOLLA McKISSOCK and WALTER *See* Walter (6)
(23) WILLIAMS *Journ Neurol Psychiat* 1941 4 107 *ibid* 1941 4 131
(24) HILL and PARR *Electroencephalography* 1950 Macdonald & Co Ltd London
(25) WEIGERT *Neurol Centralbl* 1901 20 597
(26) NORRIS *Amer Journ Cancer* 1936 27 421
(27) SAUERBRUCH quoted by SCHUMACHER and ROTH *Mitt a d Gren geb d Med u Chir* 1912 25 746
(28) BLALOCK HARVEY FORD and LILLIENTHAL *Journ Amer Med Assocn* 1941 117 1529
(29) VIETS *Brit Med Journ* 1950 i 139
(30) KEYNES *Brit Med Journ* 1940 ii 611

There is little doubt that the origin of the idea can be found in the researches of Whipple (3) and his collaborators These workers produced a severe secondary anæmia in dogs by bleeding and proceeded to study the effects of various food substances on blood regeneration They found that if liver were added in large quantities to the diet there was a remarkable increase in the blood count which appeared to be entirely due to the liver Shortly after the publication of these results Minot and Murphy (4) described the application of the same principles to the treatment of pernicious anæmia In 1926 they reported the effect of a diet rich in liver in 45 cases of pernicious anæmia These results were very striking indeed since they showed that one month s treatment increased the blood count by about 2 000 000 red cells A series of publications followed with the result that the regimen was fully upheld and now experience has proved that the blood picture can be restored almost to normal in practically all patients

Castle and his colleagues of Boston (5 6) by means of a large number of experiments on the action of healthy gastric juice on beef muscle have been able to show that the juice of the healthy stomach contains some unknown ingredient (intrinsic factor) which through its action on an extrinsic factor contained in muscle protein results in the formation of the anti anæmic or hæmopoietic substance This is able to produce a remission in pernicious anæmia equal in degree to that produced by liver extract The identity of the intrinsic factor has not been established but it is believed that it is quite unrelated to HCl or pepsin and the work of Taylor et al (7) suggests that it is probably a ferment Formijne (8) concludes that no reaction can occur *in vitro* between the intrinsic and extrinsic factors and he suggests that the reaction occurs in the intestinal wall It is thermolabile and capable of acting in a neutral medium These researches established the ætiological importance of the stomach and its secretion in the development of pernicious anæmia The anti anæmic substance thus formed is absorbed from the intestine and stored in the liver and to a lesser degree in the kidneys and spleen In pernicious anæmia the intrinsic factor is absent from the gastric juice and achylia gastrica is usually present Cohn et al (9) have shown that the active principle is neither protein lipoid nor carbohydrate but is either a nitrogenous base or a polypeptide

lead or mercury A leuco erythroblastic anæmia results from involvement of the bone marrow in carcinomatosis myelomatosis Hodgkins disease and osteosclerosis By some writers Groups 1 and 4 in the above classification are called the dyshæmopoietic anæmias

An alternative classification is based on the size of the red cells and their hæmoglobin content This is of great clinical value as it emphasises the fact that in general the macrocytic anæmias respond to liver and the hypochromic anæmias to iron therapy Iron is however required in addition to liver for macrocytic anæmias if the saturation index is low The following classification follows that of Wintrobe (2) —

1 *Macrocytic Anæmias* In this group the average size of the red cell is increased The mean corpuscular volume is greater than 94 cμ The group includes pernicious anæmia achrestic anæmia the anæmia associated with liver disorders and with myxœdema the anæmia of sprue the pernicious anæmia of pregnancy the anæmia of chylous diarrhœa and of Diphyllobothrium latum infestation

2 *Normocytic Anæmias* Here the average size of the red cell is normal The mean corpuscular volume is between 78 and 94 cμ The cells may be normochromic or hypochromic The anæmias included are aplastic anæmia the anæmia following an acute hæmorrhage and that due to malaria

3 *Simple Microcytic Anæmias* There is a slight decrease in the size of the red cells The mean corpuscular volume is less than 78 cμ The cells are normochromic Such anæmias may occur in chronic infections and in carcinoma

4 *Microcytic Hypochromic Anæmias* The red cells are smaller than normal and their hæmoglobin content is considerably reduced The mean corpuscular concentration is less than 32 / Examples of anæmias of this class are simple achlorhydric anæmia the anæmia of the Plummer Vinson syndrome the anæmia resulting from chronic hæmorrhage or from ankylostomiasis the simple nutritional anæmia of infants and chlorosis

The Macrocytic Anæmias

Pernicious Anæmia The use of liver in the treatment of pernicious anæmia constitutes a therapeutic advance of similar importance to the discovery of insulin for the treatment of diabetes

There is little doubt that the origin of the idea can be found in the researches of Whipple (3) and his collaborators These workers produced a severe secondary anæmia in dogs by bleeding and proceeded to study the effects of various food substances on blood regeneration They found that if liver were added in large quantities to the diet there was a remarkable increase in the blood count which appeared to be entirely due to the liver Shortly after the publication of these results Minot and Murphy (4) described the application of the same principles to the treatment of pernicious anæmia In 1926 they reported the effect of a diet rich in liver in 45 cases of pernicious anæmia These results were very striking indeed since they showed that one month s treatment increased the blood count by about 2 000 000 red cells A series of publications followed with the result that the regimen was fully upheld and now experience has proved that the blood picture can be restored almost to normal in practically all patients

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The Macrocytic Anæmias

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the anti anæmic principle Goldhammer (15) has shown that the volume of gastric juice is reduced in pernicious anæmia from a normal of 150 ml per hour to 20 ml per hour in pernicious anæmia The anti anæmic activity per unit volume however was the same in pernicious anæmia as in health This observation supports the view that the change in pernicious anæmia is a quantitative rather than a qualitative one

Very little can be stated with regard to the mode of action of liver extract The changes produced in the blood as the result of successful treatment include the restoration of a normal red cell count return of the colour index to normal and an increase in the number of white cells with disappearance of the relative lymphocytosis Special attention is directed to the reticulocytes present in the blood These are immature red cells normally present in the blood to the extent of 0 1 to 1 / of the red cells They cannot be seen in dried stained films but are demonstrated by vital staining in wet films A blood film is made on a slide which has previously been coated with a thin dried film of a dye such as cresyl blue A cover slip is then placed on the blood film which is examined wet (Davidson et al (16)) The reticulocytes contain a network granules or dots stained blue They probably correspond with the red cells showing polychromasia in the ordinary dry films In untreated pernicious anæmia the reticulocytes are usually somewhat increased in number as 2 / or more of the red cells In cases of pernicious anæmia treated with liver in which a good response can be expected the reticulocytes rise rapidly to 15 or 30 / in a week and then fall suddenly (reticulocyte crisis) to reach normal in about three weeks The increase in the red cell count begins usually about the time of the reticulocyte crisis and persists for several weeks after The excess of bilirubin in the serum disappears early at the time of the rise in the reticulocyte count, as shown by Dyke and Greener (17) An increase in the eosinophils after the administration of raw liver has been noted by Whitby (18) and other workers this usually reaches a maximum which may be as high as 26 / between the eighth and twelfth weeks of treatment Achlorhydria persists after liver treatment

Stomach Preparations Following the initial papers of Castle Sturgis and Isaacs (19) published their observations on the anti anæmic principle contained in dried whole hog s stomach The

The Source of the Intrinsic Factor Meulengracht (10) using the pig's stomach found that preparations from the pyloric gland region were highly active as regards the presence of the anti anæmic factor whereas those from the fundus gland region were inactive It therefore appeared probable that the intrinsic factor is connected with or produced by the pyloric glands Anti anæmic activity was also demonstrated in preparations from the duodenum and it was considered that this was due to Brunner's glands which are histologically identical with the pyloric glands The two sets of glands are called the pyloric gland organ The cells in the fundus of the stomach which produce hydrochloric acid and pepsin would not appear from these findings to be concerned with the production of the intrinsic factor It has been known for many years that there is atrophy of the gastric mucous membrane in pernicious anæmia and it therefore appeared probable that the atrophy would affect the cells which produce the intrinsic factor Almost simultaneously investigations on this point were carried out by Meulengracht (11) and by Magnus and Ungley (12) Meulengracht examined histologically the stomach and duodenum in 8 cases of pernicious anæmia and to his surprise found that although changes were present in the fundus portion with atrophy of glands and disappearance of the parietal and chief cells in all the cases the glands in the pyloric region seemed relatively well preserved and no histological changes could be demonstrated in Brunner's glands Magnus and Ungley examined the stomach from 7 cases of pernicious anæmia In all a characteristic non inflammatory atrophic lesion was found localised to the region of the body mucosa which did not affect the pyloro duodenal region The work of Fox and Castle (13) appears to explain this anomaly Extracts were made from the human stomach shortly after death and it was shown that those areas containing the fundus type of cell yielded a substance highly active in hæmopoietic properties whereas the pyloric gland organ extracts were inactive Man's stomach differs therefore in this respect from that of the pig If this is so the findings of Schenken et al (14) are contradictory They prepared liver extracts in cases of carcinoma of the stomach in man and determined their anti anæmic properties When the carcinoma involved all the stomach except the pyloric portion the liver extract was potent When the pyloric portion was affected the liver extract did not contain

The active factor is not vitamin B_1 B_4 riboflavin or nicotinic acid but it may be the extrinsic factor It is now generally agreed as Davidson (97) stated that marmite alone will not cure pernicious anæmia during the acute stage He states that many cases in the relapse stage fail to respond to vitamin B and that marmite cannot replace liver or hog s stomach extract Marmite is efficacious in the treatment of tropical macrocytic anæmia the anæmia of sprue and of cœliac disease In these diseases however there is no achlorhydria and the stomach produces the intrinsic factor which acts on the extrinsic factor to form the anti anæmic substance In those cases of pernicious anæmia in which a response to marmite is obtained there is possibly a small amount of the intrinsic factor present in the gastric juice In subacute com bined degeneration of the cord associated with pernicious anæmia marmite is not of value Berk et al (28) suggest that the extrinsic factor of Castle present in food may be closely related to vitamin B_1 and that the intrinsic factor in the gastric juice is necessary for the optimum utilisation of vitamin B_1 or of chemically related substances present in various foods Further Bethell et al (29) have shown that in cases of untreated pernicious anæmia vitamin B_{12} is found in the fæces in amounts greater than are required to treat such patients by injection of the vitamin This supports the view that pernicious anæmia results from deficient absorption of B_1

Treatment of Pernicious Anæmia

In a very severe case a preliminary transfusion of 500 ml of compatible blood given slowly is of great value if due care is taken in matching blood

Parenteral Administration of Liver In addition an intra venous injection of a liver extract such as Neo Hepatex or Hepastab Forte 5 ml should be administered slowly or 6 to 8 ml of Neo Hepatex or Hepastab given intramuscularly the first day and 2 to 4 ml on the following days until a reticulocyte response is obtained For subsequent treatment a preparation such as Neo Hepatex or Hepastab is injected intramuscularly The mainten ance dose is 2 to 4 ml weekly until the blood is normal and there are no symptoms Later 2 to 4 ml are required every two three or even six weeks

indications for its use are the same as those for liver and liver extract Accounts have been published in which stomach extract has produced a remission where liver has failed Thus Renshaw (20) describes a patient refractory to liver and liver extract but responding readily to stomach extract No definite explanation has been offered for their action but the most ingenious is that of Castle who suggests that the autolysis which occurs after death results in the interaction of the healthy gastric juice with the muscle protein of the gastric muscularis and that by this means the necessary factor is formed Wilkinson (21) showed that the mucous membrane and the muscle layer of the hog s stomach yield a light powder when desiccated at 40 C *in vacuo* Preparations from both sources are effective in the treatment of pernicious anæmia and weight for weight are equal to or better than a liver diet He also showed that normal human gastric juice when given during meals to a patient suffering from pernicious anæmia causes improvement Later Wilkinson and Klein (22) demonstrated that an active gastric extract can also be obtained by expression of the stomach juice using a pressure of 2½ to 3 tons per square inch and subsequently treating the juice with alcohol This active principle they termed hæmopoietin They regard this as the same as or closely allied to Castle s intrinsic factor Wilkinson and Klein (23) further showed that when hæmopoietin is allowed to act on beef outside the body a relatively heat resistant substance is obtained which resembles the liver active principle Wilkinson (24) considered hæmopoietin to be an enzyme like substance which is very easily destroyed by heat and which requires a substrate for its action This hæmopoietin is only found in the stomach of carnivora

Yeast Preparations The beneficial effect of marmite in the *treatment of certain megalocytic anæmias* requires consideration and the facts observed may be briefly stated Marmite is a yeast extract rich in the vitamin B complex containing the antineuritic vitamin B_1 and nicotinic acid Wills (25) in India found that marmite was as active as liver in causing regeneration of cells in the pernicious anæmia of pregnancy and in tropical macrocytic anæmia *Wills and Evans* (26) have subsequently shown that tropical macrocytic anæmia does not respond to the highly purified liver extracts which cure pernicious anæmia It does respond however to crude liver extract and to autolysed yeast extracts

Administration of Iron and Vitamin C If the mean corpuscular hæmoglobin is below 3_% iron should be given by mouth in the form of ferri et ammon cit gr 30 t i d in addition to the liver treatment Paræsthetic symptoms may be relieved by the iron In some cases of refractory pernicious anæmia as described by Wilkinson (30) ascorbic acid 100 to 150 mg must be given by mouth in addition to the liver in order to obtain a remission of the disease

Oral Administration of Stomach Preparations These are made as described above by desiccation and are not extracts Wilkinson (33) in 1931 showed that patients suffering from pernicious anæmia rapidly improved with 200 g of fresh uncooked mucous membrane of hog s stomach daily Desiccated preparations of whole hog s stomach are also available such as Pepsac or Gastrosic Twenty five grams are equivalent to 100 g of fresh whole hog s stomach The usual dose is 10 g three times a day The powder must be given cold either in soda water cold milk oxo bovril port or burgundy Wilkinson claims that this treatment is superior to liver treatment in the speed of remission the relief of gastro intestinal symptoms and in the rate at which normal health is regained Thus Wilkinson (34) has found that in seven to twelve days the reticulocytes reach a peak of 30 to 70 / followed by a rapid rise of the blood count The maintenance dose is up to 28 g of Pepsac daily The stomach treatment is considerably cheaper than liver treatment In conclusion there are three essentials in the use of stomach preparations (1) The preparation must be active (2) It must be free from micro organisms Neglect of this simple precaution has resulted in the death of the patient (3) The preparation must be given cold

Parenteral administration of stomach preparations is not possible

The Treatment of Nervous Lesions When nervous lesions such as subacute combined degeneration of the cord are present the dose of liver extract should be _ or 3 times that given for uncomplicated cases F C Wilkinson (personal communication) says that highly refined liver extracts such as Anahæmin may be used provided the batches have been tested and shown to be potent The use of vitamin B_{12} in these cases is described on p 94 Wilkinson (34) states that patients who have been bedridden owing to postero lateral sclerosis of the spinal cord have so improved

In America liver extracts are standardised in units the unit being *the amount of material which when given daily to patients* suffering from pernicious anæmia has produced satisfactory hæmopoietic response In a severe case 15 units of liver extract should be given in a single or in divided doses within the first six hours Five to 10 units should then be injected daily for a week and subsequently 10 to 15 units weekly until the blood count is normal A maintenance dose of 10 to 15 units every one to four weeks is required

In 1935 Dakin and West (30) obtained the anti anæmic principle of liver in a still more concentrated state It is known as Anahæmin and is put up in ampoules containing 100 mg in 1 ml The initial dose for a severe case of pernicious anæmia is 0 5 ml repeated daily for a week or a single intramuscular injection of 4 ml may be given Subsequently a maintenance dose of 2 ml is injected every two to four weeks Examen or Hepastab Forte may be used instead the dose being approximately double that of Anahæmin Proteolysed liver was prepared in 1943 by Davis et al (31) by digesting minced liver with papain a proteolytic enzyme which reacts at the natural pH of minced liver approximately 5 6 The product is put up as a dry powder freely soluble in cold water It is administered dissolved in warm water flavoured with pepper and salt the dose being 2 dr to 1 oz daily It has been found of value in cases of megaloblastic anæmia refractory to the parenteral administration of liver including idiopathic cases and those associated with pregnancy sprue and the puerperium

Hypersensitivity to Liver Extracts A primary reaction may occur after the first injection of a liver extract especially if a crude extract is used There may be rise of temperature shivering malaise flushing headache giddiness shortness of breath and collapse Secondary reactions may occur with later injections and are allergic in nature These include skin rashes tachycardia dyspnœa eosinophilia rigors and even death At the first sign of an allergic reaction an injection of 1 ml of adrenaline should be given followed by an antihistaminic drug such as Benadryl mg 50 or Antihsan mg 50 Subsequently treatment should be maintained by the use of a stomach preparation or by vitamin B_{12}

Folic acid must not be used in the treatment of pernicious anæmia (see p 90) The use of vitamin B_{12} in the treatment of pernicious anæmia is considered on p [illegible]

body in adequate quantities Clinically it differs from pernicious anæmia in that the age of onset is between early adult life and old age the sexes are equally affected the gastric acidity is normal or almost normal there is no disturbance of the gastro intestinal tract or central nervous system and no pyrexia or evidence of hæmolysis The liver and spleen are not usually enlarged Examination of the liver in one case showed that the anti anæmic principle was present in sufficient amount to produce a reticulocyte crisis in a patient suffering from true pernicious anæmia It was found in about 1/ of cases thought to be suffering from pernicious anæmia Achrestic anæmia has not met with universal acceptance thus some workers such as Vaughan (39) suggested that it is a megalocytic anæmia associated with liver disease and others such as Castle and Minot (39) Zanaty (41) and Davidson and Fullerton (42) have pointed out difficulties in differentiating it from aplastic anæmia Mahler and Greenberg (43) have reported a case of hyperchromic macrocytic anæmia refractory to liver extract in which they say the weight of evidence strongly favours achrestic anæmia as described by Israëls and Wilkinson Israëls and Wilkinson (44) after a further study of six new cases have strengthened their views as to the existence and nature of achrestic anæmia They say that the only changes in the liver are fatty ones associated with a severe anæmia and study of the bone marrow differentiates it from aplastic anæmia (see p 328) One case of achrestic anæmia with achlorhydria was found in 1 100 cases thought to be suffering from pernicious anæmia Bomfort and Rhoads (45) are inclined to believe that achrestic anæmia is not a specific syndrome

Treatment Life may be prolonged by repeated blood transfusions and by intensive anti anæmic treatment Liver extract 2 ml should be given intramuscularly t i d for ten to fourteen days Subsequently 4 to 6 ml should be injected weekly In addition a stomach preparation such as Pepsac 1 oz should be given daily by mouth Despite treatment the disease appears to be inevitably fatal

Macrocytic Anæmia associated with Hepatic Disorders Wintrobe and Shumacker (46) found eleven instances of macrocytic hyperchromic anæmia among 43 cases of hepatic disorders which included such conditions as cirrhosis passive congestion carcinoma and acute yellow atrophy Free hydrochloric acid was present in

with stomach preparations that they have been able to return to work

The Prognosis in Pernicious Anæmia The immediate prognosis in pernicious anæmia has been revolutionised by the introduction of liver therapy Previous to 1926 the mortality rate was nearly 100% and death occurred in periods up to two and a half years after the diagnosis was made Now it is very rare for a patient to die during the acute stage if adequate treatment is given In every case the correct maintenance dose of liver or stomach preparation must be determined and controlled by regular blood examinations The position is comparable with the administration of insulin and its control by blood sugar readings Witts (35) has emphasised the importance of adequate after treatment in cases of pernicious anæmia as every patient is under the threat of subacute combined degeneration of the cord The maintenance dose either of a liver or stomach preparation must be sufficient to keep the hæmoglobin over 90% and the red cells over 4 000 000 per c mm The blood should be examined every month The advantage of giving intramuscular injections of liver extract every two to four weeks is that the patient is compelled to remain under medical supervision Hill (36) has drawn attention to the fact that the death rate from pernicious anæmia fell substantially in 1928 following the introduction of liver treatment into England in 1927 Since 1931 there has however been a rise again in the death rate This may be due to inadequate after treatment to the disease becoming refractory to treatment after a time or to patients dying from other diseases than pernicious anæmia especially intercurrent infections i e to faulty certification Wilkinson (37) writes In January 1948 I have still alive 1 170 patients on stomach or liver therapy including 1 patient of 100 years of age 7 over 90 99 over 80 and 396 over 70 In general health they compare more than favourably with contemporary non anæmic people

Achrestic Anæmia (χρησθαι = to utilise) In 1935 Wilkinson and Israels (38) described a type of severe macrocytic anæmia which in some respects resembles pernicious anæmia but is resistant to liver therapy They considered that it is due to inability to utilise or possibly mobilise from the storage depots (such as liver kidney brain) the anti anæmic principle responsible for the correct maturation of megaloblasts although such may be present in the

Osler (51) in 1919 described a severe anæmia of pregnancy and this is of a pernicious type It is more common in India than in temperate zones and usually shows itself between the sixth and eighth months of pregnancy As described by Whitby (5) it may be plastic or hypoplastic in type In the former case there is megalocytosis with a high colour index and signs of blood regeneration but in the hypoplastic or aplastic variety there is no sign of regeneration of erythrocytes the colour index is high and there is megalocytosis or normocytosis A chlorotic type of anæmia due to deficient iron supply may also complicate pregnancy Here the colour index is low and there is no megalocytosis The pernicious anæmia of pregnancy differs from Addisonian pernicious anæmia in several respects Thus achlorhydria is usually absent and there is a tendency to spontaneous recovery after the pregnancy is terminated Clinically the patient has usually been in good health and there is no evidence of infection The patient becomes deadly pale with severe dyspnœa and often œdema or pyrexia Hæmorrhage may occur from the nose gastro intestinal tract or vagina The condition may be mistaken for an internal hæmorrhage Davidson et al (53) have investigated 16 cases of severe megalocytic anæmia occurring during pregnancy or the puerperium The colour index may be below unity Some of these were refractory to treatment by liver extract iron and yeast and in such cases life has to be maintained by blood transfusions until the hæmatinic treatment takes effect

Treatment In very severe cases blood transfusion of 500 ml should be given at short intervals followed by the daily administration of liver by mouth or injection or of stomach extract by mouth 1 oz daily This treatment should be continued until the pregnancy is terminated and the anæmia is cured

The Normocytic Anæmias

Aplastic Anæmia In this disease there is a progressive failure of function of bone marrow The anæmia is progressive without remissions The colour index is usually between 0 9 and 1 There are no reticulocytes The total white cell count is low about 1 000 and the polymorphonuclear cells are much reduced There is no achlorydria

Treatment Liver is not effective Blood transfusions are the

the gastric juice of 6 out of 10 cases examined The anæmia was not severe and remissions are likely to occur apart from treatment The authors suggest that the anæmia is possibly due to impairment of storage of the hæmopoietic principle in the liver or to a combination of partial gastric disturbance and incomplete liver damage Rosenberg (47) investigated 48 cases of cirrhosis of the liver mostly of an advanced type In 43 cases there was macrocytosis but in only 8 was the red cell count below 2 5 millions

Macrocytic Anæmias in Myxœdema These are of three types simple macrocytic normochromic Addisonian macrocytic normochromic and macrocytic or normocytic hypochromic anæmia In other cases of myxœdema a hypochromic or normochromic anæmia is found Anæmia of some type is present in about 50% of all cases of myxœdema The simple macrocytic hyperchromic anæmia can be cured by the administration of thyroid alone as was shown by Bomford (48) The anæmia is never severe but it may take several months to affect a cure The Addisonian type of anæmia required thyroid and liver extract and the hypochromic anæmia is curable with thyroid and iron

Tropical Macrocytic Anæmia The Anæmia of Sprue The Anæmia of Cœliac Disease These anæmias are probably nutritional in origin the gastric secretion being normal Response to treatment therefore will occur either with liver extract or with marmite Thus Vaughan and Hunter (49) have recorded good results with marmite in the megalocytic anæmia occurring in cœliac disease

The Nutritional Megalocytic Anæmia of Pellagra This has been studied by Moore and his co workers (50) The diets of their 25 patients had been very deficient in animal protein and the vitamin B complex Free HCl was present in the gastric juice in the majority of cases and from their experiments Moore concluded that the anæmia was not due to a deficiency of the intrinsic factor but to a prolonged lack of Castle's extrinsic factor in the diet associated in many cases with poor absorption from the intestines Macrocytic anæmia due to food deficiency can therefore occur without avitaminosis

The Pernicious Anæmia of Pregnancy Several types may occur in association with pregnancy and the puerperium Thus pregnancy may *complicate true Addisonian pernicious anæmia*

In some cases a mixture containing Acid hydrochlor dil m 60 tinc aurantii m 10 aquam ad m 120 in a glass of water t d s with meals relieves the digestive symptoms The ferri et ammon cit may produce diarrhœa in which case Blaud s pill should be substituted in doses of gr 15 t d s or Tab Fersolate one t d s

The Nutritional Anæmia of Infants Mackay and Goodfellow (61) have studied this subject in the hospital type of infant in London They estimate that 51 / of artificially fed babies and 45% of breast fed babies are anæmic (taking as the standard of anæmia a Hb figure 10 / below normal) They consider this is due to a deficiency of iron and mineral substance in the food

Treatment This should be started before the age of two months as a prophylactic measure A mixture containing Ferri et ammon cit gr 1½ aq chlorof ad m 60 can be used At first a few drops are given t d s and the dose can be increased to m 60 or 90 or more t d s as required As an alternative measure a medicated dried milk can be used containing ferri et ammon cit in such proportion that between 4 and 9 grains are given daily It was found that in the infants treated with iron the morbidity rate was halved as compared with a control series not so treated and the weight showed a definite increase in the treated cases

The Intravenous Administration of Iron

It was formerly thought that iron given by mouth is absorbed from the stomach and duodenum and excreted into the colon It is now believed that no iron is excreted into the intestine and that the excess of iron in the fæces which is noticed when iron is taken by mouth is iron which has not been absorbed Ferrous salts are more readily absorbed than are ferric ones and iron is probably absorbed in the ferrous state The iron is thought to combine with an acceptor substance a protein called apoferritin in the mucosal cells of the stomach and duodenum This results in the formation of ferritin The iron is released from ferritin to the blood when the plasma content falls below a certain level The iron combines in the plasma with β_1 globulin It has been shown that before the administration of iron there is very little ferritin or apoferritin in the mucosal cells The absorption of iron causes the formation of apoferritin and when ferritin yields its iron to the plasma the apoferritin dis appears It is possible that in the iron refractory types of anæmia

most hopeful line of treatment and the use of concentrated red cell suspensions the preparation of which is described by MacQuaide and Mollison (54) and Williams and Davie (55) avoids an undue increase of the blood volume

The Microcytic Hypochromic Anæmias

Simple Achlorhydric Anæmia (Idiopathic microcytic anæmia Essential hypochromic anæmia) This condition was described by Faber (56) in 1914 and attention has been especially directed to it in England by Witts (57) Hartfall and Witts (58) and Davies (59) and in America by Damashek (60) It affects particularly women between the ages of thirty five and fifty the chief symptoms being those of flatulent dyspepsia and dyspnœa The tongue may be smooth and red and the nails brittle or concave (koilonychia) The dietary has usually been lacking in meat and green vegetables for some time and owing to the achlorhydria iron absorption is deficient The average amount of iron in a normal diet is 10 to 20 mg of which 7 to 9 mg are available There is a daily loss of 5 to 20 mg in the fæces and 0 5 to 1 mg in the urine

The blood The red cells may number 4 millions per c mm with a hæmoglobin percentage of only 40 Frequently there is microcytosis There is no increase of bilirubin in the blood

The stomach The gastric juice contains mucus in excess and an absence of free hydrochloric acid in 70 to 80 % of cases The stomach empties rapidly or in normal time Injection of histamine provokes some secretion of hydrochloric acid in from 15 to 25 % of cases showing achlorhydria Further the gastric juice is not deficient in the intrinsic factor of Castle for on incubation of the juice with beef protein the anti anæmic principle is developed which is curative of pernicious anæmia Hartfall and Witts (58) found the intrinsic factor below normal in some cases of simple achlorhydric anæmia but it has not been conclusively shown to be completely absent in any case This probably explains why the patient does not develop pernicious anæmia although Damashek (60) suggests that the gastric juice is deficient in the power to digest organic iron

Treatment Iron should be given in large doses such as Ferri et ammon cit gr 30 aq chlorof ad fl oz 1 Fl oz 1 t d s In about six or eight weeks the hæmoglobin figure is usually normal

containing about 3% of metallic iron in a sucrose solution The saccharated oxide was found to be far less toxic than the ferric hydroxide it is however a variable compound and liable to precipitate on autoclaving Slack and Wilkinson (65) in 1949 published the results of their experiments with various iron preparations The most satisfactory was an iron sucrose solution obtained from anhydrous ferric chloride sucrose anhydrous sodium carbonate and sodium hydroxide The final solution after filtration and autoclaving was clear and dark brown with a pH of about 10 5 It contained 2% elemental iron and was given intravenously in repeated doses to over 120 people It was found that the total dose of elemental iron required in iron deficiency anæmia is 24 5 mg for each 1% deficit of hæmoglobin as determined on the Haldane scale plus 50% to restore the iron reserve in the tissues to normal Alternatively 100 mg of elemental iron should increase the hæmoglobin by 0 55 g per 100 ml The plan of dosage adopted was 25 mg on the first day 50 mg on the second day 100 mg on the third day and 200 mg on the fourth and subsequent days It is not wise to give a larger single dose than 200 mg of this preparation The solution used contained 2% elemental iron with 200 mg in 10 ml A No 2 stainless steel Record needle was used The rate of injection should be 2 ml a minute and the same vein should not be used on consecutive days To avoid local thrombophlebitis no pressure should be made over the vein when the needle is withdrawn and movements of the arm should be made to encourage the rapid removal of the iron from the site of injection Ferrivenin is another form of iron sucrose preparation used by these workers Most of the 60 cases of iron deficiency anæmia were treated as out patients and only one developed a mild reaction at the 200 mg dose Ten of these patients had proved refractory to full doses of iron given by mouth It was noted that reticulocyte peak may reach 10% to 18% in seven to ten days from the beginning of the treatment In almost all of the cases there was a rapid symptomatic and hæmatological response to treatment and unless chronic infection was present, nearly 100% of the injected iron appeared to be utilised Thus a woman suffering from rheumatoid arthritis with a calculated iron deficiency of 1550 mg required a total of 550 mg in three separate courses before there was a satisfactory response

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Some patients suffering from iron deficiency anæmia do not respond to the oral administration of iron even though they tolerate it others are unable to take iron by mouth owing to gastro intestinal disturbances To meet the requirements of these two classes of patients various experiments and investigations have been made to produce a preparation which can be safely administered intravenously in doses sufficiently large to cause hæmoglobin regeneration The B P injectio ferri contains ferric chloride The maximum dose for intramuscular injection is 30 minims and this is equivalent to gr 1/10 of iron or gr ¾ of iron and ammonium citrate Such a small dose is insufficient to relieve iron deficiency anæmia

Various ionised iron compounds such as ferric ammonium citrate have been used for parenteral administration but when given intramuscularly they are painful and when injected intravenously they are poisonous in amounts larger than 10 to 20 mg of elemental iron

Cappell (62) in 1930 investigating the distribution of iron in the tissues and its subsequent fate showed that a 10 / solution of saccharated oxide of iron in amounts up to 0 3 ml per 20 g body weight was well tolerated by mice on intravenous injection There was no fear of producing hæmochromatosis as the iron was not deposited in the pancreas A considerable interval elapsed before these findings were applied clinically

Goetsch and his colleagues (63) in 1946 injected colloidal ferric hydroxide or colloidal ferric oxide intravenously into 8 patients suffering from hypochromic microcytic anæmia A single massive injection varying from 0 608 g to 1 32 g was used The reticulocyte response was higher than would be expected in oral therapy and there was good hæmoglobin regeneration Severe toxic reactions however occurred in all but 2 cases which contra indicated the use of this measure as a therapeutic procedure The toxic reactions are largely due to precipitation of iron in the blood leading to pulmonary and in some cases to systemic emboli There may also be gastro intestinal disturbances and depression of the nervous system resulting in coma and death Nissim (64) a year later tried the intravenous injection of colloidal ferric hydroxide and saccharated oxide of iron The latter was a B P C preparation

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end between the syringe and needle A little blood withdrawn from the vein can be clearly seen in the glass connection The blood serves as a buffer for the iron solution The injection must be made slowly about 2 ml a minute as rapid injection appears to cause venospasm If the needle becomes displaced from the lumen of the vein during the injection the skin around is quickly stained a slate grey colour The injection must then be stopped immediately The iron solution must never be mixed with saline as an unstable preparation may be produced If it is wished to administer the iron slowly by drip transfusion as in a case of a child in which it has been necessary to cut down on the vein the calculated amount of the iron solution is added to 900 ml of 5 / dextrose solution and run in at the rate of 30 to 45 drops a minute In this way 35 or 40 ml of iron solution (700–800 mg iron) may be run in during the course of eight or nine hours

Agranulocytic Angina and Malignant Neutropenia

The term agranulocytic angina was applied by Schultz (70) in 192? to describe the illness of patients exhibiting severe progressive oral sepsis with marked prostration and neutropenia Schilling (71) in 19.9 suggested the alternative name of malignant neutropenia Two varieties of acute cases are now described—primary agranulocytic angina and secondary malignant neutropenia In the former occasionally without any known cause the patient is taken ill with fever severe prostration and ulcerative lesions in the mouth or other mucous membranes such as the vagina There is a total reduction of the white cells often to below 1 000 per c mm and a very low polymorphonuclear count Many of these cases are now thought to be due to amidopyrine or to an amidopyrine and barbiturate compound to which the patient has become susceptible by a preliminary sensitising dose

Plum (72) in Copenhagen recorded seven cases of agranulocytosis due to amidopyrine and since 1933 many cases of agranulocytosis have been recorded developing after the therapeutic administration of amidopyrine preparations such as Pyramidon Gardan and Amidophen or to barbiturates containing amidopyrine such as Allonal Cibalgin Compral Somnosal Veramon and Veropyron Agranulocytosis has also followed the administration of dinitrophenol dinitrocresol benzol arsenic preparations Sedormid

Govan and Scott (66) injected Ferrivenin in 25 cases of the anæmia of pregnancy and found that about 40 mg of elemental iron were required to raise the hæmoglobin by 1% The larger amount of iron required may be due to the fœtus using some of the iron The response to treatment was rapid, but in one case there was a severe reaction with faintness giddiness, a sensation of bursting in the head vomiting slow pulse and extra systoles The attack passed off in a few minutes but no further injections of iron were given in this case It is possible that the reaction was due to the fact that there had been an interval of 12 days between the previous injection and the one which proved toxic The rapid response enables the treatment to be given during the last few weeks of pregnancy for it is at this time that the anæmia becomes severe The necessity for a blood transfusion is thus avoided Nissim and Robson (67) also in 1949 published their work on the methods of preparation and standardisation of saccharated iron oxide using mice as test animals They claimed that their preparation of iron sucrose was less toxic than Ferrivenin and that a dose of 300 mg was not followed by reactions A preparation of this nature is available under the name of Ivíron 100 mg of iron being contained in 5 ml Impurities in the sugar used in preparation may cause a severe anginal like pain in the chest arms and back Sinclair and Duthie (68) used Ferrivenin in a series of cases of hypochromic anæmia associated with rheumatoid arthritis A test dose of 50 mg was injected followed by 4 daily doses of 200 mg Poor responses were obtained in those cases in which the sedimentation rate of the red cells was high Ramsey (69) using Ferrivenin tried to see if it were possible to give the amount of iron required 300 to 800 mg in a single intravenous injection This somewhat bold experiment was quickly followed by a severe toxic reaction in the 11 cases so treated The toxic reaction could be avoided by giving an initial dose of 100 mg which is slowly increased to 200 or 300 mg

The dose we are accustomed to employ is 50 mg on the first day 100 mg on the second and third days and 200 mg on the subsequent days until the calculated required dose is given Owing to the dark colour of the iron solution it is not possible to see whether the needle is in the lumen of the vein by withdrawing a little blood into the syringe The difficulty can be overcome by using a short glass connection known as a sightfeed with a metal adaptor at each

may appear worse during the first days of the treatment but the improvement is usually definitely noted by the fourth or fifth day and the blood count returns to normal by the eighth or tenth day In addition to the injections a careful search should be made for ulceration of the mucous membranes or local abscesses elsewhere and these should be appropriately treated Blood transfusions are of doubtful value but if the patient is intolerant of pentnucleotide as shown by symptoms of cardiac distress after a trial dose a small transfusion (500 ml) should be given The use of penicillin in the treatment of agranulocytosis is described in Chapter II

Results Jackson and his co workers (80) have reported sixty nine cases treated by pentnucleotide by themselves and other physicians with a recovery rate of 74% In cases treated previously by various other methods a cure was only effected in 25 % of cases Cases successfully treated in England by pentnucleotide have been recorded by Bulmer (81) Marriott (82) Wilkinson and Israels (83) and by Smith (84) and we have also treated a severe case of malignant neutropenia with good results Fairley and Scott (85) report a fatal case of agranulocytic angina in which there was a complete absence of response to nucleotide K 96 injections It must however be remembered that the American observers report a recovery rate of only 74 % Israëls and Wilkinson (86) say that pentnucleotide has little effect in chronic neutropenia For the latter Gupta and Watts (87) recommend the intramuscular injection of liver It should be noted that pentnucleotide is useless in the treatment of aplastic anæmia of leukæmia or of sepsis without neutropenia and leucopenia We have also successfully treated with penicillin a case of agranulocytosis due to thiouracil

Sternal Puncture

Ghedini (88) of Genoa in 1908 introduced the method of human bone marrow biopsy by puncturing the shaft of the tibia and withdrawing a small amount of marrow for microscopical examination Seyfarth (89) in 1923 recommended that the sternum should be trephined rather than the tibia and Arinkin (90) in 1927 advised that a thick needle should be used for sternal puncture Peabody (91) in 1927 investigated bone marrow obtained by trephine from the tibia in cases of pernicious anæmia Salah (92) in 1937 described the needle which now forms the model of that

bismuth and gold salt. A fatal case following the administration of *prontosil flavum* has been recorded in Holland by Borst (73) and in America death has followed the use of sulfanilamide as recorded by Schwartz and his co workers (74) and by Berg and his co workers (75) It has also resulted from the use of other sulphonamide drugs especially if improperly given in small doses for prolonged periods or in amounts totalling more than 25 to 30 g and from the use of thiouracil Both types of disease if untreated in the majority of cases pursue a rapidly fatal course The blood changes in malignant neutropenia resemble those of agranulocytic angina but they are secondary to some infection such as pneumonia sinusitis osteomyelitis staphylococcal septicæmia or liver abscess Further chronic cases of neutropenia have been described by Doan (76) in which the white cells from time to time fall below 4 000 per c mm owing to a reduction of the granulocytes with accompanying symptoms of ill health and a liability to septic infections Barsby and Close (77) describe a case of a recurrent agranulocytosis in which the symptoms recurred for three to seven days after the menstrual periods for four years and during the last seven months of the patient s life were accompanied by a neutrophil agranulocytosis

Treatment In 1924 Jackson (78) demonstrated the existence in normal human blood of pentose nucleotides Jackson and his co workers (79) in 1931 and 193- reported the effect of treatment of cases of agranulocytic angina and malignant neutropenia with a preparation called nucleotide K 96 This is now available as pentnucleotide and is put up in ampoules of 10 ml containing 0 7 g

In very acute cases 0 7 g of pentnucleotide in 100 ml of saline is injected slowly intravenously every morning for four days and 0 7 g of pentnucleotide in 10 ml of distilled water is injected in the evening intramuscularly The intramuscular injections are continued subsequently twice daily until the white cell count has definitely risen and then an injection is given once a day until the white count has been essentially normal for three consecutive days If there is myocardial degeneration the intravenous injection should not be given owing to risk of heart failure from the reaction of dyspnœa and palpitations which is often provoked In the average case only the daily intramuscular injections are given The patient

may appear worse during the first days of the treatment but the improvement is usually definitely noted by the fourth or fifth day and the blood count returns to normal by the eighth or tenth day In addition to the injections a careful search should be made for ulceration of the mucous membranes or local abscesses elsewhere and these should be appropriately treated Blood transfusions are of doubtful value but if the patient is intolerant of pentnucleotide as shown by symptoms of cardiac distress after a trial dose a small transfusion (500 ml) should be given The use of penicillin in the treatment of agranulocytosis is described in Chapter II

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used for sternal puncture He used a lumbar puncture needle (*made of hard steel*) with its stillet (*sic*) cut to 3 cms length its point sharpened but made broad so that it will not break A movable shield is made to fit round the needle with a screw to fix it at the required distance

The figures for the normal range of marrow cells have been worked out by Nordenson (93) Young and Osgood (94) Vogel Erf and Rosenthal (95) and by Hynes (96) In some cases especially in aplastic anæmia and myelosclerosis it is necessary to know the histological structure of bone marrow which can be done by removing a disc of bone with a 1 cm trephine

Method A Salah or Klima pattern needle is sterilised in hot oil The patient is *given an injection of morphine gr* ¼ *or aspirin gr* 10 and nepenthe m 15 by mouth half an hour before the operation *The skin over the manubrium sterni is sterilised with alcohol or* iodine The skin subcutaneous tissues and periosteum are infiltrated with about *1 ml of* 2% procaine hydrochloride *solution in* the mid line at the level of the third costal cartilage using a fine intradermic needle with a short bevel The patient lies on his back with a pillow between the shoulders The Salah needle is then passed through the skin down to the sternum The adjustable stop is fixed 5 mm above the skin and the needle is forced through the outer table of the bone by means of gentle taps with a small hammer When it is felt to enter the medullary cavity the stylet is withdrawn and a dry 1 ml syringe is attached Marrow juice is

TABLE I

Cells	Neutrophil per cent	Eosinophil per cent	Basophil per cent
Segmented polymorphs	9 –30	0–3	0–0 5
Non segmented polymorphs	0 –40	0–2	0–1
Metamyelocytes	[illegible] 12	0–2 5	—
Myelocytes	[illegible] 8	0–1	—
Premyelocytes	0 5 5		
Myelo blasts	0 – 5		
Lymphocytes	[illegible] 0	Total cell count 25 000	
Plasma cells	0 – 1	to 100 000 per c mm.	
Monocytes	0 5		
Normoblasts	[illegible] 15	Myeloid-erythroblastic	
Erythroblasts	[illegible]	ratio	
Megaloblast	0 4	[illegible] 1 to 8 1	

then sucked into the syringe This produces a sensation of discomfort or even of actual pain Hynes (96) recommends that 0 25 ml of marrow fluid be withdrawn and placed in a tube containing Wintrobe s dry oxalate mixture The tube is prepared by placing in it 0 1 ml of a solution of 0 2 / potassium oxalate and 0 8 / ammonium oxalate and drying it in the incubator Smears are then prepared stained by Leishman s method and a differential count is made of 400 or 500 cells

Dangers Death from perforation of the right ventricle during sternal puncture has been recorded by Scherer and Howe (97) and by Bardhan (98) In Scherer and Howe s case the needle penetrated the sternum to a depth of ¾ in and in Bardhan s 2 cases the screw guard was fixed at a level of 8 mm and 11 mm Hæmophilia is usually regarded as a contra indication although successful cases have been recorded by Limarzi et al (99)

Normal Marrow Findings

The normal ranges in the bone marrow differential count as described by Hynes (96) are shown in Table I The cells in the lymphocyte series include the lymphoblast when it is possible to distinguish it from the myeloblast and a variety of intermediate forms The first six types of cell in the table are myeloid in nature and the last three are erythroblastic

Abnormal Marrow Findings

Leukæmia Sternal puncture is of great value in the diagnosis of aleukæmic (leucopenic) leukæmia The marrow changes here are similar to those found in leukæmia with leucocytosis

The Acute Leukæmias The marrow is very cellular 70 to 99% of the marrow cells being primitive white cells either of the myeloid or lymphatic series The proportion of primitive red cells is low but the erythroblasts almost equal in number the normoblasts There is no premature hæmoglobinisation of the erythroblasts such as occurs in the bone marrow in pernicious anæmia This maturation defect is probably associated with the macrocytic anæmia which is so often present in leukæmia

Chronic Lymphatic Leukæmia Here 40 to 90 / of the marrow cells are lymphocytes which may be normal small lymphocytes or

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Myelocytes	[illegible] – 8	0–1	—
Premyelocytes	0 5 5		
Myeloblasts	0 – 2 5		
Lymphocytes	[illegible] –20	Total cell count 25 000 to 100 000 per c mm	
Plasma cells	0 – 1		
Monocytes	0 – [illegible]		
Normoblasts	[illegible]	Myeloid-erythroblastic ratio	
Erythroblasts	[illegible]		
Megaloblasts	0 – 4	[illegible] 1 to 8 1	

then sucked into the syringe This produces a sensation of discomfort or even of actual pain Hynes (96) recommends that 0 25 ml of marrow fluid be withdrawn and placed in a tube containing Wintrobe s dry oxalate mixture The tube is prepared by placing in it 0 1 ml of a solution of 0 2% potassium oxalate and 0 3 / ammonium oxalate and drying it in the incubator Smears are then prepared stained by Leishman s method and a differential count is made of 400 or 500 cells

Dangers Death from perforation of the right ventricle during sternal puncture has been recorded by Scherer and Howe (97) and by Bardhan (98) In Scherer and Howe s case the needle penetrated the sternum to a depth of ¾ in and in Bardhan s 2 cases the screw guard was fixed at a level of 8 mm and 11 mm Hæmophilia is usually regarded as a contra indication although successful cases have been recorded by Limarzi et al (99)

Normal Marrow Findings

The normal ranges in the bone marrow differential count as described by Hynes (96) are shown in Table I The cells in the lymphocyte series include the lymphoblast when it is possible to distinguish it from the myeloblast and a variety of intermediate forms The first six types of cell in the table are myeloid in nature and the last three are erythroblastic

Abnormal Marrow Findings

Leukæmia Sternal puncture is of great value in the diagnosis of aleukæmic (leucopenic) leukæmia The marrow changes here are similar to those found in leukæmia with leucocytosis

The Acute Leukæmias The marrow is very cellular 70 to 99 % of the marrow cells being primitive white cells either of the myeloid or lymphatic series The proportion of primitive red cells is low but the erythroblasts almost equal in number the normoblasts There is no premature hæmoglobinisation of the erythroblasts such as occurs in the bone marrow in pernicious anæmia This maturation defect is probably associated with the macrocytic anæmia which is so often present in leukæmia

Chronic Lymphatic Leukæmia Here 40 to 90 / of the marrow cells are lymphocytes which may be normal small lymphocytes or

show abnormalities in the size and structure of the nucleus The myeloid cells and the erythroblasts show a maturation defect

Chronic Myeloid Leukæmia The marrow in early cases shows an increase in myeloblasts premyelocytes and myelocytes Lymphocytes and monocytes are few or absent There is a maturation defect in the nucleated red cells In the final stages of the disease the marrow resembles that found in acute myeloid leukæmia

Pernicious Anæmia In untreated cases 25 to 45% of the marrow cells are megaloblasts and erythroblasts and premature hæmoglobinisation is present The polymorphs are greatly decreased but the number of earlier myeloid cells is not affected

Aplastic Anæmia The bone marrow should be studied by histological section and not by needle puncture The marrow is usually hypoplastic showing a lack of mature cells particularly of the myeloid type and an increase of primitive cells resembling small lymphocytes In other cases the marrow is hyperplastic with many primitive cells Israels and Wilkinson (100) state that *megaloblasts are not found*

Achrestic Anæmia Israëls and Wilkinson (100) found megaloblastic hyperplasia resembling that of pernicious anæmia Hynes (96) examined the marrow in one case and recorded an increase in the early hæmoglobinised erythroblasts and also in the normoblasts The megaloblasts were normal Mahler and Greenberg (101) found increased megaloblastic activity with diminished numbers of normoblasts

Other Macrocytic Anæmias In sprue and in carcinoma of the stomach marrow changes are found similar to those in pernicious anæmia

Secondary Anæmias Young and Osgood (94) demonstrated an erythroblastic reaction in the bone marrow in the anæmia which accompanies hæmorrhage toxæmias and infections

Polycythæmia In some cases there is an increase in the erythroblasts and myeloid cells in the marrow In others the only change found is an increase in the segmented polymorphs

Acholuric Jaundice In chronic cases the marrow is usually normal in the acute phases however there is an increase of the normoblasts erythroblasts and megaloblasts

Agranulocytosis In some cases there appears to be a

maturation defect the bone marrow having its normal number of cells but all the myeloid cells being myeloblasts In others there is an aplasia of the marrow lymphocytes and plasma cells being the only white cells present

Gaucher s Disease The typical Gaucher s cells are present in the marrow

Kala azar The Leishman Donovan bodies are seen in the monocytes in the marrow

Malaria Parasites are more numerous than in the blood being seen in the red cells and leucocytes in the marrow

Myelomatosis An excess of plasma cells may be present in the marrow

Carcinomatosis In secondary carcinoma of bone carcinoma cells in groups or isolated are found in some instances in the marrow

The appearances of the bone marrow in the abnormal conditions described above are largely based on the article of Hynes (96)

Conclusions Sternal puncture is chiefly of value in the diagnosis of leukæmia with leucopenia enabling a confident opinion to be expressed which cannot be done by blood examinations alone It is also of value in the diagnosis of obscure cases of anæmia and certain other diseases as described above

Bone Marrow Transfusion

In some cases it is extremely difficult if not impossible to introduce fluids into the body by the direct intravenous route This is likely to be so in adults if the veins are collapsed owing to shock or if they are compressed by œdema of the subcutaneous tissues Similar difficulties are met with in young children profoundly dehydrated as the result of gastro enteritis and in infants whose veins are either too small to receive the cannula or too tender to retain it even if successfully introduced

That fluid may pass from the bone marrow cavity through the emissary veins into the general circulation was demonstrated by Tocantins and O Neill (102) in America in 1936 Experimenting on the transplantation of marrow from animal to animal they observed that if two needles were inserted into the marrow cavity of the rabbit s femur one at either end of the bone and 5 ml of saline injected through the proximal needle only about 2 ml of fluid would come up through the needle at the distal end Further

show abnormalities in the size and structure of the nucleus The myeloid cells and the erythroblasts show a maturation defect

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syringe half filled with 3 8 % sodium citrate solution is attached to the cannula a little citrate solution injected into the marrow cavity and on slowly withdrawing the piston the syringe is seen to fill with the red bone marrow Some more citrate is now injected the syringe disconnected and the cannula joined to the transfusion apparatus the tubing of which must be free from air bubbles The wings of the cannula and the tubing are held in position by strapping attached to the chest wall and the transfusion allowed to proceed The container holding the fluid should be between 4 and 6 feet above the level of the sternum if the pressure is too high pain will ensue The flow of blood can be aided by oxygen pressure or by the use of a blood saline mixture

Tibial Transfusion This is the method of choice for infants and young children The leg must be maintained in a semi externally rotated position by means of a splint the other leg being loosely tied to the side of the cot to prevent dislocation of the cannula by kicking Gunz and Dean (105) found that the legs were most satisfactorily controlled by firmly bandaging the soles of the feet to a padded splint the feet being about 9 inches apart and the ends of the splint tied to the sides of the cot Gimson (106) has devised four needles and stylets of different length and size for tibial transfusion in infants and young children (see Fig 34) The flange is fixed and each needle has its own stylet The needles measure below the flange ¼ inch ⅜ inch

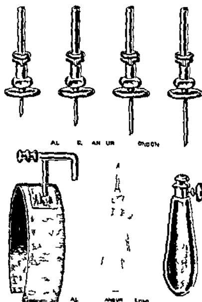


Fig 34 Needle etc for bone marrow transfusion

investigations proved that the fluid passed direct from the bone marrow into the general circulation Tocantins and O Neill were not slow in applying their experimental results to clinical medicine and using the bone marrow of the sternum the lower end of the femur and the upper end of the tibia successfully infused citrated blood citrated plasma 5% dextrose solution and physiological saline into the general circulation both of adults and of children These workers considered that the method was likely to prove of value in patients suffering from extensive burns mutilating wounds generalised œdema circulatory collapse or where the veins are poorly developed or have been rendered useless by repeated punctures or by injections of hypertonic saline A case of diabetic coma was successfully treated by infusion of insulin and dextrose solution into the bone marrow attempts at intravenous injection having failed as the patient was pulseless and the veins were collapsed

Hamilton Bailey (103) in 1944 reported on his experiences of the method in England and concluded that not only is the medullary cavity of the sternum as good a receptor as a vein for all kinds of infusions and for Pentothal to induce anæsthesia but it is particularly valuable for the resuscitation of the very shocked and the very young Again in 1946 (104) he recorded how a patient was rapidly revived after a severe surgical operation by the injection of over a pint of saline into the sternal marrow cavity the infusion being completed within three minutes using a sternal puncture trocar and cannula and Record syringes

Sternal Transfusion Strict aseptic precautions must be taken to prevent the likelihood of osteomyelitis developing as a complication In the method described by Hamilton Bailey (103) the patient lies flat and 2 ml of 1 / procaine hydrochloride solution are infiltrated into the skin subcutaneous tissues and periosteum in the midline just above the manubrio gladiolar junction the site of the injection then being massaged with a sterile swab so that the bony landmark can be clearly felt The special trocar and cannula for sternal puncture is now pushed with a boring movement through the outer plate of the bone just above the manubrio gladiolar junction and directly the outer plate is felt to be penetrated the direction of the instrument is changed the point being directed upwards towards the patient s head and the wings now prevent the point from piercing the inner plate of the bone The trocar is withdrawn and a Record

Massey (108) has reviewed the literature of bone marrow infusions and concludes that the intra tibial method is very satisfactory for the administration of parental fluids in infants when difficulty is found with the intravenous method

Heparin

McLean (109) in 1916 extracted from dog liver a substance which retards the coagulation of blood *in vitro* Further investigations were carried out by Howell and Holt (110) in 1918 and the anti coagulant substance was called heparin In 1923 Howell and Holt (111) described an improved method for its preparation and defined the unit of heparin as being that weight of substance which would prevent 1 ml of cat blood from clotting for twenty four hours when kept in the cold McHenry and Glaister at Best s suggestion subsequently prepared small quantities of heparin from beef liver and Charles and Scott (112) in 1933 evolved a process by which it could be prepared in much larger quantities from liver They also obtained a blood anti coagulant presumably heparin from skeletal muscle and lung In 1936 the same workers (113) prepared a crystalline barium salt of heparin having a high potency of about 500 units per mg They gave the empirical formula of heparin as $C_{25}H_{65}O_{50}N_2S_5$ By treating the crystalline barium salt with excess of ammonium carbonate solution at 65 C the barium was removed the heparin was then precipitated by acetic acid centrifuged down washed with ether and dried For use a solution is prepared to which tricresol is added and it is sterilised by passing through a Berkefeld filter Jorpes and Bergstrom (114) classified heparin as a mucoitin polysulphuric acid closely related to chondroitin sulphuric acid Best (115) has suggested as a standard that the unit is the activity contained in 0 01 mg (10γ) of the barium free material *i e* 100 units in 1 mg There is evidence to suggest that heparin is either formed in or stored in the mast cells as had been shown by Jorpes Holmgren and Wilander (116)

Mode of Action It is believed by Howell that heparin is normally present in the blood Howell and Holt (110) showed that heparin has no effect in preventing coagulation when added to mixtures of thrombin and fibrinogen and differs in this respect from hirudin They suggested that it delays coagulation by inhibiting the activation of prothrombin to thrombin Mellanby

½ inch and ⅝ inch The two shorter needles are size 18 S W G the two longer ones 16 S W G The ¼ inch needle is for premature infants and the newborn the ½ inch and ⅝ inch needles for children of about 5 years but the size of the child and the amount of subcutaneous fat *must be taken into consideration in selecting the* needle The handle (see Fig 54) is screwed on to the stylet and the needle and stylet inserted after preliminary cleaning of the skin and anæsthetisation with procaine into the subcutaneous surface of the tibia and at right angles to the subcutaneous bony plate The site selected is inferior and medial to the anterior tibial tuberosity below the epiphyseal line and above the nutrient artery The flange rests on the skin the point of the needle being in the marrow cavity The stylet is withdrawn and a small amount of marrow wells up or is sucked up into a syringe partly filled with saline attached to the needle A little saline is injected into the marrow cavity to make sure that the needle is in position The adaptor (see Fig 54) filled with transfusion fluid and connected to the tubing from the dripper and bottle from which air bubbles have been excluded is fitted to the needle and the transfusion begun A screw clip on the tubing can be used to adjust the rate of flow and in one case Gimson maintained the transfusion for as long as six days Owing to the difficulty in judging the size of needle required Behr (107) prefers for tibial transfusion a modification of the Hamilton Bailey trocar and cannula furnished with adjustable wings Hartmann s solution is valuable as a transfusion for infants suffering from dehydration due to gastro enteritis as it maintains the acid base equilibrium It contains sodium chloride 6 g potassium chloride 0 4 g calcium chloride 0 2 g lactic acid 2 4 ml N/10 NaOH q s to neutralise the lactic acid and water to 1 000 ml There may be difficulty in transfusing blood if undiluted even if the container is raised to as much as 7 feet above the needle and Gunz and Dean (105) considered the method unsuitable for the transfusion of blood For dehydrated infants Behr (107) recommends the transfusion of half strength Hartmann s solution in 5% dextrose the rate of flow being varied according to the weight of the patient For infants weighing less than 5 lbs the flow should be 4 drops a minute if under 7½ lbs 6 drops a minute under 10 lbs 8 drops a minute and under 15 lbs 12 drops a minute The drops apply to a drip bulb delivering 1 ml in 16 drops

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(117) considered that heparin acts as an antithrombase and says it is improbable that heparin is responsible for the normal fluidity of blood He suggests that heparin is concerned with the prevention of localised thrombosis in the immediate neighbourhood of disintegrating tissue Best (118) in 1939 upheld the view that heparin apparently combines with the antithrombin of the serum albumin fraction and thus acts as an anti coagulant by increasing the affinity of this substance for thrombin

Clinical Applications Heparin has been used in surgery in medicine and in blood examinations *Heparin is available in sterile* solution in rubber capped vials of 8 ml ($\frac{1}{2}\%$ = 5 mg per ml) of 5 ml (1% = 10 mg per ml) and of 5 ml (5% = 50 mg per ml) This preparation contains 500 units per mg Heparinised tubes (10 ml) each containing 1 mg are available for collection of blood samples In man 1 mg of heparin per kg body weight when given intravenously prolongs the coagulation time to approximately forty minutes For clinical use the average dose for an adult is 75 to 150 mg given intravenously in the form of a 5% solution This dose is repeated every third or fourth hour four or five times a day and the effect should be checked by determination of the coagulation rate which should be kept at about fifteen to twenty minutes by the Lee and White method (normal four to seven minutes)

Surgical Murray et al (119) after showing that heparin will prevent thrombosis *resulting from mechanical and chemical injury* to blood vessels in dogs first administered heparin to a patient in a surgical ward of the Toronto General Hospital in 1935 The solution was injected into the brachial artery under local anæsthesia and samples of blood from the veins of the injected arm showed a rise in clotting time from the normal of six minutes to that of eighteen minutes Subsequently heparin was administered post operatively as a prophylactic measure against thrombosis The injections were commenced two to three hours after the operation and were continued for twenty four to 120 hours An effort was was made to keep the clotting time at fifteen to twenty minutes The operations included appendicectomy herniotomy resection of colon etc Heparin has also been used in arteriotomies for embolism Crafoord (120) advises that in these cases it may be necessary to start heparinisation during the operation When heparin has thus been

given or when hæmorrhage unexpectedly sets in during the course of heparin treatment the original coagulation time of the blood may be restored by the injection of protamine Chargaff and Olson (121) described this neutralising effect on heparin and Jorpes et al (122) state that 60 mg of protamine (Clupein) instantaneously neutralise the effect of 100 mg of heparin They say however that the protamine injection should only be used as a last resort, and a blood tranfusion will usually effect the necessary hæmostasis Richmond (123) records a case which shows that the intravenous injection of heparin to prevent post operative thrombosis is not devoid of danger An intravenous injection of 20 mg of heparin in normal saline was given to a girl aged eleven years after removal of a twisted ovarian dermoid Massive hæmaturia occurred sixteen hours later the blood coagulation time being seventeen minutes The patient was very collapsed but recovered with bladder irrigations and a blood transfusion

Medical *Ploman (124) used heparin in the treatment of acute* thrombosis of the central vein of the retina Intravenous injections of a 5 / heparin solution were given with a fine needle Four in jections were made daily the first three of 50 mg and the last of 100 mg Marked improvement or full restoration of vision was obtained Magnusson (125) recorded a case of thrombosis of the posterior inferior cerebellar artery in a woman aged forty eight Intravenous injections of a 5 / solution were used 150 mg in the morning and afternoon of the first day and the same amount three times a day for five days He states that the case was a severe one and that improvement was more rapid than would be expected normally Kelson and White (126) have reported some favourable results in the treatment of subacute bacterial endo carditis by the combined use of heparin and sulphapyridine Their method is as follows The contents of a 10 ml vial of heparin (10 000 units) are added to 500 ml of normal saline and the solution is given by continuous intravenous drip for fourteen days The rate of flow usually 15 to 25 drops per minute is regulated to maintain as well as possible the venous clotting time at approximately one hour Sulphapyridine g 4–6 daily is given by mouth for a week before during and a week after the heparin treatment Blood transfusions are given if there is an anæmia of 3 5 million red cells or lower All patients are saturated with

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ascorbic acid 200 mg being given by mouth four times a day for three days and subsequently 100 mg a day

We have used this method in 2 cases of subacute bacterial (streptococcus viridans) endocarditis without benefit the patients dying a few weeks later If the sulphapyridine which is given before the heparin fails to sterilise the blood there appears to be an increased risk of the occurrence of vascular accidents such as cerebral hæmorrhage McLean Meyer and Griffith (127) reviewing 67 cases treated by this method report very unfavourably on it.

Heparin has also been used in the *treatment of thrombophlebitis* including the migratory type in pulmonary embolism and in coronary thrombosis Murray (128) states that in phlebitis the effect of heparin is most noticeable when given early in the disease In the migratory type although the heparin will probably relieve the symptoms of thrombosis it will not affect the cause of the disease In an average case of thrombophlebitis heparin is given for ten days After the first three to four days the patient is encouraged to move actively in bed and he should get up by the tenth to twelfth day Murray (128) records that 22 cases of massive pulmonary embolism have been treated with heparin with pleasing results no patient died from the embolism and dyspnœa and distress were rapidly relieved Some cases of coronary thrombosis have been treated but experimental results indicate that heparin may prevent the occurrence of mural thrombosis with subsequent risk of peripheral embolus formation Stansfield (129) treated successfully a case of puerperal cerebral thrombophlebitis with heparin using an intravenous drip of 5% dextrose saline containing 200 mg heparin in each pint The coagulation time was prolonged to forty minutes by this treatment Other cases of recovery have however been recorded without the use of heparin Massive hæmaturia has resulted from the treatment of a case of cavernous sinus thrombosis with sulphathiazole and heparin as described by Ershler and Blaisdell (130) The evidence given in the article indicates that the bleeding was due to the heparin and not to the sulphathiazole

Blood Transfusions Hedenius (131) in 1937 reported on the use of heparin in blood transfusion A 5 / solution is used 1 mg per kg body weight being injected intravenously into the donor The blood coagulation time is prolonged rapidly at first and returns

to normal in one and a half hours Ten minutes after the injection blood is removed from the donor with any standard equipment and the whole blood in its natural state injected into the recipient If syringes are used the plunger should be moistened with a drop of sterile paraffin to prevent cell destruction and kinase production The coagulation rate of the recipient is not affected The technique employed in citrate transfusions may also be used heparin being added to the drawn blood instead of citrate using 20 mg heparin (0 4 ml of a 5% solution diluted with 10 ml sterile saline) for 500 ml of blood These methods have also been reported on favourably by Sappington (132) in America

Blood Examinations Heparinised blood may be used for biochemical and cytological examinations but is not suitable for performing the Wassermann reaction The blood is placed in a 10 ml tube containing 1 mg of heparin which is distributed as a smear on the inside of the tube A leucocyte count should be carried out within two hours of collecting the blood

Conclusions It is evident that the administration of heparin is not devoid of danger from cerebral or renal hæmorrhage and its use seems only to be justified in cases in which the treatment is very carefully controlled

Plasma Transfusion

Clinically many occasions arise where plasma is preferable to whole blood for transfusion They occur where there is marked fluid loss but no loss of red cells as in post operative shock and in shock following burns and wounds It may also be used to restore blood volume after a hæmorrhage should no blood be available In burn shock with marked hæmoconcentration the hæmatocrit reading is a useful guide in treatment Infusion of plasma until the plasma–cell ratio is normal and constant greatly reduced the mortality

Whilst plasma has the advantage of being easily and quickly administered to any patient without matching and requires no special storage the risk of transmitting the virus which produces serum hepatitis with the possible complication of liver necrosis and cirrhosis must be borne in mind (See Chapter VI) At first large pool plasma batches (*i e* at least 300 donors) were used with varying reports for the incidence of serum hepatitis In the series

of Brightman and Korns (133) it was 4 5% and Havens (134) reported it as high as 16% It is now recommended that only small pool batches (i e not more than 10 donors) should be used when the incidence falls to a rate comparable with that of whole blood transfusion In the series of Lehane and others (135) the incidence *from small pools was 1 3% and from whole blood 0 8%*

The early claims that irradiation with ultra violet light would destroy the virus have been disproved by the reports of Rosenthal Bassen and Michael (136) James Korns and Wright (137) and others

In 1943 Gronwall and Ingelman (138) in Sweden began work on a plasma substitute named Dextran which is now available It is a *non antigenic polysaccharide built entirely of glucose molecules* a 6% solution having a viscosity similar to that of plasma It is metabolised within five days of administration and carries no risk of transmitting serum hepatitis

Dextran produces very heavy rouleaux formation Therefore blood samples required for grouping and matching should be taken before the administration of Dextran

The Rhesus Blood Group System

In 1940 Landsteiner and Wiener (139) showed that the antibody produced by the immunisation of rabbits with the blood of the *Macacus rhesus monkey agglutinated the red cells of 85% of* the white population of New York Those persons whose cells are agglutinated are termed Rh positive and the remainder Rh negative Wiener and Peters (140) described antibodies which reacted in the same way as the anti Rh rabbit sera in 3 patients who developed transfusion reactions from the administration of homologous ABO blood In 1941 Levine Katzin Burnham and Vogel (141) demonstrated that erythroblastosis fœtalis is due to rhesus incompatibility between mother and child

Further antibodies agglutinating 80 / 70% and 30% of test cells were discovered and in 1943 Fisher (142) suggested that the rhesus system consisted of 3 pairs of allelomorphic genes which he called C and c *D and d E and e Allelomorphic genes are alternative* genes capable of occupying the same locus on a chromosome These 3 allelomorphic pairs give rise to eight phenotypes

Fisher's notation	*Convenient shorthand notation*
CDe	R_1
cDE	R_2
cDe	R_0
Cde	R
cdE	R
CDE	R_z
CdE	R_y
cde	r

Further alleles some very rare have since been described increasing the number of Rh phenotypes Three alleles of C and c are C^w reported by Callender and Race (143) and C and c by Race Sanger and Lawler (144) Stratton (145) described D an allele of D and d Recently two alleles of E and e were reported E by Armytage Ceppellini Ikin and Mourant (146) and e by Gilbey (147)

However from the clinician s point of view the important factor in most cases is whether or not the patient is D positive or D negative

Production of Rhesus Antibodies These antibodies are immune and rarely if ever occur naturally They occur either from stimulation due to multiple pregnancies or from repeated blood transfusions During pregnancy some of the fœtal red cells escape into the mother s circulation via the placenta and should they carry an Rh antigen derived from the father but not possessed by the mother they may act as a foreign substance sensitise the mother and cause the production of antibodies in her serum Generally the first pregnancy sensitises the mother and antibodies appear in future pregnancies These antibodies return to the fœtus via the placenta destroy fœtal red cells and produce erythroblastosis fœtalis Similarly transfusion of blood of heterologous Rh group may sensitise the patient with the production of antibodies at subsequent transfusions As small a volume as 1 or 2 ml of blood may be sufficient to sensitise even if given in early childhood

Obviously blood of a different Rh type to that of the patient is frequently transfused and many mothers carry a fœtus of another Rh type to that of themselves and yet antibody production is much

of Brightman and Korns (133) it was 4·5% and Havens (134) reported it as high as 16%. It is now recommended that only small pool batches (*i.e.* not more than 10 donors) should be used when the incidence falls to a rate comparable with that of whole blood transfusion. In the series of Lehane and others (135) the incidence from small pools was 1·3% and from whole blood 0·8%.

The early claims that irradiation with ultra-violet light would destroy the virus have been disproved by the reports of Rosenthal, Bassen and Michael (136), James, Korns and Wright (137) and others.

In 1943 Gronwall and Ingelman (138) in Sweden began work on a plasma substitute named Dextran which is now available. It is a non-antigenic polysaccharide built entirely of glucose molecules, a 6% solution having a viscosity similar to that of plasma. It is metabolised within five days of administration and carries no risk of transmitting serum hepatitis.

Dextran produces very heavy rouleaux formation. Therefore blood samples required for grouping and matching should be taken before the administration of Dextran.

The Rhesus Blood Group System

In 1940 Landsteiner and Wiener (139) showed that the antibody produced by the immunisation of rabbits with the blood of the Macacus rhesus monkey agglutinated the red cells of 85% of the white population of New York. Those persons whose cells are agglutinated are termed Rh positive and the remainder Rh negative. Wiener and Peters (140) described antibodies which reacted in the same way as the anti-Rh rabbit sera in 3 patients who developed transfusion reactions from the administration of homologous ABO blood. In 1941 Levine, Katzin, Burnham and Vogel (141) demonstrated that erythroblastosis fœtalis is due to rhesus incompatibility between mother and child.

Further antibodies agglutinating 80%, 70% and 30% of test cells were discovered and in 1943 Fisher (142) suggested that the rhesus system consisted of 3 pairs of allelomorphic genes which he called C and c, D and d, E and e. Allelomorphic genes are alternative genes capable of occupying the same locus on a chromosome. These 3 allelomorphic pairs give rise to eight phenotypes

example an Rh negative mother who has produced anti D should the husband be homozygous D positive all future children will almost certainly be affected but if the husband is heterozygous D positive only half the children may be affected

How is it possible for a child to suffer from erythroblastosis when both parents have been reported as Rh positive? In routine work because the full genotyping sera are very rare and as D is the most powerful antigen blood is examined for the presence or absence of the D antigen only with anti D serum Therefore a mother (R_1R_1) (CDe/CDe) and father (R_2r) (cDE/cde) would both be reported Rh positive but the mother might develop anti E or anti c as she lacks both these antigens while the fœtus will carry either or both of them

Anthropological Value of Blood Group Systems The blood group frequencies vary in races throughout the world and are valuable in anthropological studies As people migrate and inter marry with local populations frequency differences tend to merge but small racially pure communities may still be found The Hungarian gipsies are known to have come from India many generations ago and a study of their ABO groups helps to confirm this Their frequency for A and B is very close to that of the Indian Hindus but quite different from that of the ordinary Hungarian population

Group B increases in frequency towards the East In Great Britain it has a frequency of 8% in Eastern Europe Poles 20% Russians 23% and in the Pekin Chinese it is 35%

Even in Britain the frequencies show significant variations Travelling from Southern England to Northern England the value for O steadily increases and that for A decreases The Icelandic people have an ABO frequency similar to that found in Northern Scotland suggesting a common ancestry Changes occur also in the Rhesus distribution The Basques have the highest Rh negative frequency known while the North American Indians and Chinese are almost entirely Rh positive The latter are said to develop erythroblastosis fœtalis only very rarely

ABO Grouping To each of two tubes one containing a volume of anti A serum and the other a volume of anti B serum add a volume of a 2% saline suspension of the washed unknown cells Similarly add a volume of the unknown serum to each of two tubes

less frequent than might be expected The explanation is that the genes have varying antigenic potency *i e* varying ability to produce an antibody if introduced into the circulation D is by far the most powerful antigen followed by C and the great majority of Rh antibodies are anti D or anti C + D E and c are weaker antigens and antibodies to either are not often encountered while e and d are very weak antigens and only two examples of anti d have been reported Thus erythroblastosis mainly affects infants of Rh negative mothers and seldom those of Rh positive mothers For the same reason it is important that Rh negative persons particularly young girls and women in the childbearing period should be transfused with Rh negative blood only

People vary in their ability to produce antibodies and this may explain why only 1 in 10 or 15 of Rh negative women married to Rh positive men have infants affected by erythroblastosis fœtalis In Great Britain approximately 1 in every 200 infants at birth have their red cells sensitised by maternal antibodies not all are clinically affected a positive direct Coombs test on the infant's red cells at birth sometimes being the only evidence of sensitisation *in vivo*

In assessing the prognosis for future pregnancies in a family where a child has suffered from erythroblastosis it is an advantage to know the Rh genotype of the husband Taking as a common

(1)
CDe/cDE cde/cde
CDe/cde CDe/cde cDE/cde CDe/cde cDE/cde

(2)
CDe/cde cde/cde
CDe/cde CDe/cde cde/cde CDe/cde cde/cde

□ Normal male ■ erythroblastosis (male)
○ Normal female ● erythroblastosis (female)

(1) Homozygous D positive husband CDe/cDE
(2) Heterozygous D positive husband CDe/cde

agglutinate in the presence of Coombs reagent and is valuable in testing newborn children suspected of suffering from erthroblastosis fœtalis and in cases of acquired hæmolytic anæmia Coombs reagent (rabbit anti human globulin serum) can be prepared by immunising a rabbit with serum from a group O person or with purified globulins

To perform the test the red cells are washed 3 times in saline to remove any trace of plasma proteins and then mixed on a slide with the Coombs reagent Should any antibody (globulin) be attached to the cell envelope agglutination will occur

Indirect The indirect test will detect incomplete antibody in a patient s serum The unknown serum is incubated with cells of known group at 37 C for one hour or more The cells are then washed three times and mixed with Coombs reagent as in the direct test Both in the direct and indirect test red cells of known type must be tested concurrently to act as controls

Detection of Antibody Samples of blood from persons requiring transfusion and who have had previous transfusions or borne children and from pregnant women who have an obstetric history suggestive of hæmolytic disease should be examined for the presence of immune antibodies Their serum should be incubated at 37 C with cells of known Rh type suspended in saline and albumin and an indirect Coombs test performed By noting the reactions with cells of known genotype the particular type of anti body may be identified The identification of the rarer antibodies can generally be done only in pecial laboratories

Matching Blood for Transfusion Donor s cells mixed on a slide with the patient s serum is not a satisfactory test of com patibility for transfusion purposes This method will not detect immune antibodies in most cases as they normally react at 37 C and are often incomplete (albumin agglutinating)

The patient s serum should be incubated at 37 C for one hour in tubes with the donor s cells suspended both in saline and albumin This method will detect any incompatibility and it is most important that such a method be used in all cases of trans fusion

Only in cases of the direst emergency where any delay in trans fusing may endanger the life of the patient is it permissible to use blood without matching and in these cases blood known to be

one containing a volume of known A cells and the other a volume of known B cells Tap the tubes to mix the contents and leave at room temperature (about 20 C) for one to two hours The tests may then be read macroscopically and if apparently negative microscopically by transferring the contents on to a glass slide Known A B and O cells must be added to tubes containing anti A and anti B to act as controls for the tests

Determination of Rh groups A volume of the unknown cell suspension is added to two tubes each containing a volume of *different anti D sera* as some D positive red cells will not react with all anti D sera The tubes are incubated at 37°C for two hours and then the cells are gently transferred on to a glass slide and read microscopically Rh agglutinates are fragile and rough handling will break them up and cause errors Both D positive and D negative red cells must be used as controls This method gives the most accurate results but two rapid methods are described That of Chown (148) requires the mixing of a high titre anti D serum with the unknown cells in their own serum in a capillary tube and incubating at 37 C for fifteen to twenty minutes In the method of Diamond and Abelson (149) a drop of high titre anti D serum is mixed with test cells in their own serum on a slide which is warmed and rocked on a special box for three minutes Not all anti D sera are suitable for these two rapid methods

Rhesus Antibodies These antibodies are immune (*i e* result from iso immunisation) and in common with other immune blood group antibodies react most strongly at 37 C Two forms exist—complete and incomplete

Complete antibodies are saline agglutinating antibodies *i e* a serum containing a complete antibody will agglutinate red cells carrying the appropriate antigen when they are suspended in saline Incomplete antibodies are also termed blocking antibodies or albumin agglutinating antibodies An incomplete antibody will not agglutinate appropriate red cells suspended in saline but will so affect them that they can no longer be agglutinated by a saline agglutinating antibody *i e* the cells are blocked However incomplete antibodies will agglutinate appropriate red cells if they are suspended in 20 / bovine albumin

Coombs Test *Direct* This test depends on the presence of an antibody on the red cell receptors and the fact that such cells will

agglutinate in the presence of Coombs reagent and is valuable in testing newborn children suspected of suffering from erthroblastosis fœtalis and in cases of acquired hæmolytic anæmia Coombs reagent (rabbit anti human globulin serum) can be prepared by immunising a rabbit with serum from a group O person or with purified globulins

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Coombs Test. *Direct.* This test depends on the presence of an antibody on the red cell receptors and the fact that such cells will

rejected A bottle of blood opened and not used within twelve hours should be discarded as bacteria may have been introduced

Paternity Testing In English law it is only possible to use blood grouping tests to exclude paternity and Race and Sanger (150) have shown that about 60 / of men wrongfully accused of paternity could be exonerated provided the tests could be carried out against all the blood group systems for which anti sera are available

Secretion The A B H and rarely O antigens are secreted in various body fluids particularly saliva of some persons and this ability to secrete is inherited as a Medelian dominant character Grubb and Morgan (151) showed that about 7" / of the London population are secretors Grubb (152) observed a close relationship between secretion and the Lewis blood groups Briefly Lewis positive persons are non secretors of A B or H substances and the majority of Lewis negative persons are secretors of A B and H

MNS Blood Group System In 19?7 Landsteiner and Levine (153 154) described the M and N antigens which give rise to three genotypes MM MN and NN An antibody now called anti S was discovered in 1947 by Walsh and Montgomery (155) S is closely associated with M and N and increases the genotypes to 6 MMS MsMs MNS MsNs NNS and NsNs (s denotes cells which do not react with anti S)

Anti M and anti N occur both as natural and immune antibodies while anti S sera are usually immune They rarely are the cause of hæmolytic reactions Anti s has so far not been demonstrated

Blood group system	First reported	Frequency in Britain		Nature of antibody	Cause of haemolytic action
P	Landsteiner and Levine 19?? (154)	P ??	+ve	usually natural	very rarely
Kell	Coombs Mourant and Race 1946 (1 6)	K 10	+ve	immune	occasionally
Lewis	Mourant 1946 (157)	Le ??	+ve	usually natural	very rarely
Lutheran	Callender and Race 1946 (158)	Lu ?5	+ve	immune	very rarely
Duffy	Cutbush, Mollison and Parkin 19 0 (1 9)	Fy 6	+ve	immune	very rarely

group O Rh negative should be used unless the patient has been previously grouped

Blood Transfusion Hazards (1) *Pyrogens* Pyrogens may cause mild pyrexia rigors and backache These reactions can be reduced to a minimum by the use of pyrogen free water for solutions and by scrupulous aseptic technique in use and cleaning of materials

(2) *Overloading the Circulation* Great care is essential in the transfusion of patients with cardiovascular disease and/or chronic anæmia A rising pulse rate and falling blood pressure, together with a persistent rise in venous pressure may herald the onset of pulmonary œdema or congestive cardiac failure Generally this type of patient should be slowly transfused with small volumes of packed red cells

(3) *Incompatible Transfusions* These may be due to the following —

(*a*) Presence of cold agglutinins
(*b*) Mistaking rouleaux formation for agglutination
(*c*) Failure to cross group with patient s serum
(*d*) Errors in grouping technique or interpretation
(*e*) Clerical errors in laboratory or ward
(*f*) Failure to carry out a satisfactory matching test

The first three pitfalls will be avoided if the cells to be tested are washed in saline and the grouping done as described above

If the matching of the donor s cells and recipient s serum is carried out at 37 C with saline and albumin as suspending fluids and when necessary with the use of the indirect Coombs test then incompatible transfusions rarely occur

Homologous blood should always be used Very rarely a person of group O may have an extremely high anti A titre and if this blood should be transfused to a person of group A a reaction may well occur The indiscriminate use of group O blood throws an unnecessarily heavy strain on group O blood donors

(4) *Transmittible Disease* The virus causing serum hepatitis is the greatest danger and Lehane and others (135) reported 0 8% as the incidence following whole blood transfusion It is possible also to transmit both syphilis and malaria by blood transfusion Donors who have had any of these diseases should be

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Further Blood Group Systems Five other blood group systems are very briefly described in the table on the previous page All are weak antigens and so rarely stimulate antibody production Any antibody which they may stimulate can be detected by the use of *saline and albumin as suspending fluids for the red cells* except in the case of the Duffy antibodies the majority of which can be demonstrated only by the use of the indirect Coombs test

Kidd System This blood group system was described in 1951 by Allen et al (160) It was discovered during the investigation of a maternal serum the child of the mother having suffered from typical erythroblastosis fœtalis 77 % of Americans were shown to be Kidd positive (Jk +ve)

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CHAPTER XIII

BIOCHEMICAL METHODS

THE essential requirements of methods used for routine analyses are accuracy speed and ease of operation Generally colorimetric methods are used as satisfying these conditions for certain types of analysis however volumetric or gravimetric procedures or the use of special apparatus are recommended

COLORIMETRY

Photoelectric colorimeters are now generally used instead of direct vision colorimeters as they have a wider field of application and eliminate the personal error there is very little to choose between the different commercial makes of single cell machines It is essential however to use a double cell null point instrument for those analyses where it is inconvenient or not possible to prepare a standard for each analysis and where a calibration curve must be used

THE COLLECTION AND STORAGE OF BLOOD SPECIMENS

The first prerequisite of an accurate blood analysis is a representative blood sample The collection of blood by capillary puncture is not to be recommended except in skilled hands because of the difficulty of avoiding adulteration of the blood by tissue fluid *On the other hand venepuncture affords a convenient method* of withdrawing blood samples ample in volume for any *series* of estimations

The technique of blood withdrawal by venepuncture is a matter of common knowledge in the medical field Not so well known however are the limitations set by chemical considerations factors which sometimes overlooked by the uninitiated can vitiate the most careful analysis

A most frequent source of error lies in the choice of anticoagulant or indeed in making a decision whether an anticoagulant should be

used at all For example fluoride a specific enzyme inhibitor is useless for any estimation involving the use or assay of an enzyme (*e g* urease in urea estimations or for the estimation of phosphatase activity) Paradoxically it is just this property of fluoride which makes it pre eminently suitable for the collection of blood samples for inorganic phosphate determination where failure to inactivate the blood phosphatase will lead to the liberation on standing of phosphate from organically bound phosphorus yielding a falsely high result Fluoride is best known for its use to prevent glycolysis where a sugar has to be estimated

Potassium oxalate is probably the most effective common anti coagulant fluoride is commonly used mixed with oxalate as fluoride alone is a poor anticoagulant For obvious reasons both these anticoagulants potassium oxalate or sodium fluoride are quite unsuitable for use when sodium or potassium themselves are being estimated and since oxalate or fluoride precipitate calcium they also invalidate this analysis

For certain analyses namely chloride bicarbonate and blood gases it is essential that the specimen be collected under liquid paraffin to avoid loss of gas from the sample and consequent ionic shift

As far as possible the tendency nowadays is to use serum for as many analyses as possible for the use of any solid anticoagulant will alter the water distribution between cells and pla ma and wherever there is any choice serum is to be used

Haemolysed specimens should always be treated with reserve especially when estimating the serum or plasma content of any substance known to be present in much higher concentration in the erythocytes *e g* potassium or the phosphatases

For the collection of specimens where anticoagulant is required a suitable concentration of potassium oxalate is 2 mg /ml of final sample and where fluoride is desired this is added as well as the oxalate to a concentration of 0 7 mg /ml Suitable concentrated solutions of these substances may be prepared and the appropriate amount of solution placed into a bottle the amount calculated for an average sample of 10 ml of blood The solution is run over the sides of the bottle which is then dried in an oven at a temperature not exceeding 110 C leaving the anticoagulant distributed in a finely divided easily soluble form

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A most frequent source of error lies in the choice of anticoagulant or indeed in making a decision whether an anticoagulant should be

B Urine

(Values as g /24 hrs unless otherwise stated)

Calcium	0 1–0 5
Chloride (as Cl)	6–9
Corticoids	1–4 mg /24 hrs
Creatine	0–0 05
Creatinine	1 0–2 0
Diastase (amylase)	2–50 units/ml
17 Ketosteroids (total neutral)	
Males	15–25 mg /24 hrs
Females post menopause	4–10 mg /24 hrs
premenopause	6–18 mg /24 hrs
Phosphate (as P)	0 5–2 0
Potassium (as K)	1 5–4 0
Pregnanediol	
Males and females (follicular phase)	0–0 5 mg /24 hrs
Females (luteal phase)	1–10 mg /24 hrs
Sodium (as Na)	2–6
Urea	10–40
Uric acid	0 1–2 0
Vitamin C	0–80 mg /24 hrs

C Fæces

Total fat	10–25 of dried fæces
Split fat fatty acid	3–10 of dried fæces
soaps	3–10 of dried fæces
Unsplit (neutral fat)	3–8 of dried fæces
Trypsin	200–2000 units per g wet fæces
Urobilinogen (= stercobilinogen)	0 05–0 3 g /24 hrs

D C S F (Values as mg /100 ml)

	Lumbar	Ventricular
Chloride (as NaCl)	720–760	720–760
Protein	10–40	5–15
Sugar (as glucose)	40–80	50–100

Flame Photometry

Flame photometry is an analytical technique now being applied to clinical biochemistry The principles of flame photometry are (1) certain elements on suitable excitation emit light of characteristic wavelength (2) the intensity of the light emitted is

Physiological Normals

(By methods described in text)

A Blood

(Values as mg /100 ml unless otherwise stated)

	Whole Blood	Plasma or Serum
Alkali Reserve (as HCO_3)	—	24-3[illegible] mEq/l
Amino acid nitrogen	4 8	3-7
Bilirubin (Van den Bergh)	—	0 1-0 8
Calcium (total)	5-[illegible]	9-11
Calcium (diffusible)	—	4 2-[illegible] 6
β Carotene	—	0 0[illegible]-0 3
Chloride (as Cl)	[illegible]-87 mEq/l	96-10[illegible] mEq/l
	([illegible]0-310)	(340-370)
Cholesterol (total)	110-230	140-280
Creatine	2-8	0 2-0 8
Creatinine	0 5-2 5	0 [illegible]-2 [illegible]
Iron	—	0 08-0 18
Liver Function tests		
Colloidal gold	—	0-1 units
Thymol turbidity	—	0-4 units
Oxygen Males		
arterial	15-24 ml /100 ml	—
venous	11-17 ml /100 ml	—
Females 2 ml	less in each case	
Phosphatase (acid)	—	1-3 K-A units/100 ml
Phosphatase (alkaline)	—	3-13 K-A units/100 ml
Phosphate (inorganic) (as P)	2 [illegible]-[illegible] 0	2-4 5
(as HPO)	(1 [illegible]- [illegible]9)	(1 2-2 6 mEq/l)
Potassium (as K)	38-64 mEq l	4 4-5 6 mEq/l
Protein total	—	[illegible]-8 0 g /100 ml
Albumin	—	3 [illegible]-6 0 g /100 ml
Globulin	—	1 5-3 0 g /100 ml
A/G Ratio	—	1 5 1-3 [illegible] 1
Fibrinogen	—	0 2-0 4 g /100 ml
Prothrombin time	—	14 -16
Concentration	—	70 / -140 /
Sodium (as Na)	74-98 mEq l	139-150 mEq/l
Sugar (as glucose)		
Total fasting		
venous or capillar	[illegible]0-110	60-110
after a meal		
venous	up to 1[illegible]0	up to 1[illegible]0
capillary	up to 180	—
Urea	1[illegible]-40	1[illegible]-40
Uric acid	1-4	1 [illegible] [illegible]

B Urine

(Values as g./24 hrs. unless otherwise stated.)

Calcium	0·1–0·5
Chloride (as Cl.)	6–9
Corticoids	1–4 mg./24 hrs.
Creatine	0–0·05
Creatinine	1·0–2·0
Diastase (amylase)	2–50 units ml.
17 Ketosteroids (total neutral)	
Males	15–25 mg./24 hrs.
Females post menopause	4–10 mg./24 hrs.
premenopause	*6–18 mg./24 hrs.*
Phosphate (as P)	0·5–2·0
Potassium (as K.)	1·5–4·0
Pregnanediol	
Males and females (follicular phase)	0–0·5 mg./24 hrs.
Females (luteal phase)	1–10 mg./24 hrs.
Sodium (as Na.)	2–6
Urea	10–40
Uric acid	0·1–2·0
Vitamin C	0–80 mg./24 hrs.

C Fæces

Total fat	10–25 % of dried fæces
Split fat fatty acid	3–10 % of dried fæces
soaps	3–10 % of dried fæces
Unsplit (neutral fat)	3–8 % of dried fæces
Trypsin	200–2000 units per g. wet fæces
Urobilinogen (= stercobilinogen)	0·05–0·3 g./24 hrs.

D C.S.F. (Values as mg./100 ml.)

	Lumbar	Ventricular
Chloride (as NaCl)	720–760	720–760
Protein	10–40	5–15
Sugar (as glucose)	40–80	50–100

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	Whole Blood	Plasma or Serum
Alkali Reserve (as HCO_3)	—	24–35 mEq/l
Amino acid nitrogen	4–8	3–7
Bilirubin (Van den Bergh)	—	0 1–0 8
Calcium (total)	5–7	9–11
Calcium (diffusible)	—	4 2–5 [illegible]
β Carotene	—	0 05–0 3
Chloride (as Cl)	76–87 mEq/l (270–310)	96–105 mEq/l (340–370)
Cholesterol (total)	110–230	140–280
Creatine	[illegible]–8	0 2–0 8
Creatinine	0 5–2 5	0 [illegible]
Iron	—	0 08–0 18
Liver Function tests		
Colloidal gold	—	0–1 units
Thymol turbidity	—	0–4 units
Oxygen Males		
arterial	1[illegible]–24 ml /100 ml	—
venous	11–17 ml /100 ml	—
Females 2 ml	less in each case	
Phosphatase (acid)	—	1–3 K–A units/100 ml
Phosphatase (alkaline)	—	3–13 K–A units/100 ml
Phosphate (inorganic) (as P)	2 5 5 0	3–4 5
(as HPO_4)	(1 5–2 [illegible])	(1 2–2 6 mEq/l)
Potassium (as K)	38–[illegible]4 mEq/l	4 4–5 6 mEq/l
Protein total	—	[illegible]–8 0 g /100 ml
Albumin	—	3 5–6 0 g /100 ml
Globulin	—	1 5–3 0 g /100 ml
A/G Ratio	—	1 5 1–3 5 1
Fibrinogen	—	0 2–0 4 g /100 ml
Prothrombin time	—	14 –16
Concentration	—	70 / –140
Sodium (as Na)	74 98 mEq/l	139–150 mEq/l
Sugar (as glucose)		
Total fasting venous or capillary	60–110	60–110
after a meal venous	up to 150	up to 150
capillary	up to 180	—
Urea	15 40	15–40
Uric acid	1–4	1 5 6

B Urine

(Values as g /24 hrs unless otherwise stated)

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Vitamin C	0–80 mg /24 hrs

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Protein	10–40	5–15
Sugar (as glucose)	40–80	50–100

Flame Photometry

Flame photometry is an analytical technique now being applied to clinical biochemistry The principles of flame photometry are (1) certain elements on suitable excitation emit light of characteristic wavelength (2) the intensity of the light emitted is

proportional to the concentration of the appropriate element in the material tested By means of an appropriately designed burner atomiser system operating under controlled conditions it is possible to provide stable and reproducible means of excitation and consequently of estimation of certain metals (see Fig 55)

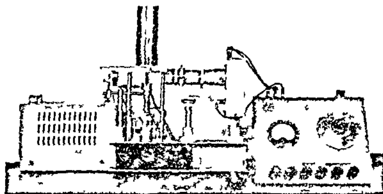


FIG 55 The Flame Photometer

The Courtauld Institute Flame Photometer was constructed for the routine estimations of sodium and potassium in biological fluids

The aspirator situated at the centre of the burner draws the solution from the beaker atomises it in the stream of oxygen and passes it into the flame The intensity of the light produced is measured by means of emission type photo cells appropriate filters being used to separate the sodium and potassium radiations A bridge amplifier system modified from that described by Bills et al (1) is employed to measure the photo cell current

The burner atomiser system used is a modified form of that of Weichselbaum and Varney (2) with the novel feature that it may be completely dismantled for cleaning being constructed from only two pieces of metal Other features include the elimination of back firing (troublesome in the original design) and injection mixing of the fuel (butane and oxygen)

In routine use the photometer illustrated gives readings consistent to ± 1 5 / The range of concentration in the aspirated solution normally employed for sodium estimations is 0·004–0·4 mEq/l and for potassium 0 002–0·2 mEq/l. At these concentrations the calibration is for practical purposes linear The final volume of the material used is limited by the size of the beakers convenient to handle (2 ml. fluid in a 5 ml beaker)

The layout of the instrument is as follows The various optical components burner and photo cell compartments are mounted on an optical bench. The compartment on the left houses the pressure gauges and needle valves that indicate and regulate the pressure of the fuel and oxygen. The burner on the left is water cooled (as is the chimney) The optical system and filter slide are mounted on the right of the instrument and on the extreme right is the photo cell compartment carrying two photo cells one for potassium and one for sodium These cells are mounted side by side and are easily interchangeable. Output from the photo cell is led by screened cables to the amplifier On the extreme left is the stabilised power supply The photo cell current is measured by the large calibrated dial which is attached to a potentiometer A detailed description of the working of this instrument is not given here because it is not commercially available and different makes of instruments would naturally have to be differently operated For those laboratories not possessing a flame photometer a suitable method for the estimation of serum sodium is that of McCance and Shipp (3) and for the estimation of serum potassium that of Jacobs and Hoffman (4) which may be suitably modified for the estimation of urinary sodium and potassium

Gasometric Analysis

Alkali Reserve Estimation (macro-method)

Method of Peters and Van Slyke (5)

Reagents (1) *Lactic acid* 1N Take one volume of lactic acid S G 1·20 and dilute with nine volumes of distilled water Place into a flask and draw a stream of air through the solution for at least one hour The incoming air should be freed from carbon dioxide by means of a soda lime tube

(2) *Lactic acid* 0 1*N* Dilute the above solution ten times with distilled water and again aerate

Method The blood is collected into a plain tube and covered with a layer of liquid paraffin If an urgent estimation is required *it is permissible to use an oxalate bottle and carry out the analysis on plasma* Upon receiving the specimen at once centrifuge and transfer the serum to a lusteroid plastic test tube For the analysis the sample must be equilibrated with carbon dioxide at the concentration present in alveolar air To do this slowly blow through the serum a stream of air containing 5% carbon dioxide (this mixture can be obtained commercially in cylinders)

Whilst the blood is being gassed *inspect the apparatus and test for leaks* To test for leaks run about 10–15 ml of distilled water into the shaking chamber seal the top cock of the chamber with mercury lower the mercury level in the chamber to the 50 ml mark and shake for three minutes to remove the air from the water Raise the water level to the 2 ml mark and read the level of the mercury in the manometer tube Re lower the mercury level again shake for one minute raise to the 2 ml mark and read the manometer Repeat this process three or four times If there are no leakages then a constant reading will be obtained a slight difference between the first two readings is permissible

For the analysis a drop of caprylic alcohol is placed into the cup and the reservoir placed in position 1 Open cocks C and D Allow the mercury to rise into the cup Close cocks D and C Place reservoir in position 2 Open cock C Slowly open cock D until the caprylic alcohol is in the capillary of the stopcock D Close D raise reservoir to position 1 Close C Lower reservoir to position 2 Measure 1 5 ml of 0 1N lactic acid into the cup 1 ml of gassed serum is drawn up into a volumetric 1 ml pipette with the tip protected by a rubber cuff and the pipette is inserted into the cup as shown in Fig 56 (Method 2) When the pipette is in position open cock D and let the serum flow into the chamber by turning cock C (Cock C *is used instead of cock D as the flow of mercury* is more easily controlled than the flow of serum) The delivery is continued until the serum has entirely left the pipette and a minute bubble of air has followed the serum into the capillary at the bottom of the cup Close cock D The bubble should float to the top if this does not happen then remove the bubble using a piece

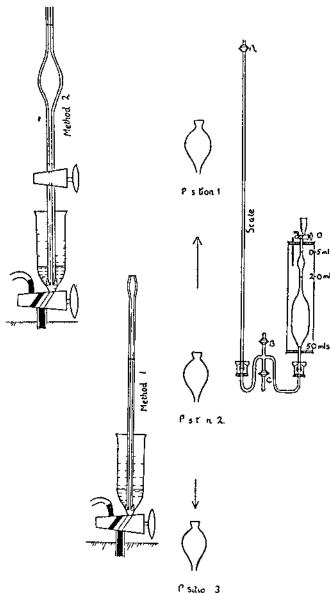


FIG [illegible]6 The Van Slyke Apparatus

(2) *Lactic acid* 0 1*N* Dilute the above solution ten times with distilled water and again aerate

Method The blood is collected into a plain tube and covered with a layer of liquid paraffin If an urgent estimation is required it is permissible to use an oxalate bottle and carry out the analysis on plasma Upon receiving the specimen at once centrifuge and transfer the serum to a lusteroid plastic test tube For the analysis the sample must be equilibrated with carbon dioxide at the concentration present in alveolar air To do this slowly blow through the serum a stream of air containing 5% carbon dioxide (this mixture can be obtained commercially in cylinders)

Whilst the blood is being gassed inspect the apparatus and test for leaks To test for leaks run about 10–15 ml of distilled water into the shaking chamber seal the top cock of the chamber with mercury lower the mercury level in the chamber to the 50 ml mark and shake for three minutes to remove the air from the water Raise the water level to the 2 ml mark and read the level of the mercury in the manometer tube Re lower the mercury level again shake for one minute raise to the 2 ml mark and read the manometer Repeat this process three or four times If there are no leakages then a constant reading will be obtained a slight difference between the first two readings is permissible

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of mercury in the cup Slowly open cock D and fill the capillary of the cock with mercury Close cock D Lower the reservoir to position 3 and close cock C when the mercury reaches the 50 ml mark Return the reservoir to position 2 Shake the chamber for two minutes at a speed of three movements per second After shaking allow the fluid to drain then slowly open cock C and by careful control allow the fluid level to rest on the 2 ml mark Read the gas pressure on manometer scale ($= P_1$) Open cock C and bring the reservoir level with the fluid in the chamber in order to bring the gas pressure to slightly above atmospheric Close cock C and open cock D if the previous manipulation has been correctly performed no movement of the gas should occur Raise the reservoir to position I By the cautious adjustment of cock C expel the gas through cock D and fill the cock with fluid Place some mercury in the cup fill the cock with mercury by lowering the reservoir and close cock D Lower the reservoir to position 3 and close cock C when the fluid level is exactly at the 2 0 ml mark Read the gas pressure on the manometer ($= P_0$) Take the temperature of the water jacket $P_1 - P_0$ = corrected pressure Read Vols CO_2 on the nomogram of corrected pressure against temperature (see Fig 57)

Alkali Reserve Estimation (micro method)

Reagent (1) 0 01*N* *Lactic acid* Dilute 0 1N lactic acid ten times with distilled water and aerate as in the macro method

Method The serum or plasma is gassed with 5 / carbon dioxide in air exactly as for the macro method For the analysis a drop of octyl alcohol and 0 ?0 ml of the saturated serum or plasma are run into the chamber of the apparatus followed by enough 0 01N lactic acid to bring the volume of the solution in the chamber down to the 2 0 ml mark The same analytical technique is used as in the macro method the reading of the pressure of the extracted gases being made at 0 5 ml volume The pressure observed is applied to the nomogram as in calculating the results of the macro determination and the final result is multiplied by 1 6 to give the correct capacity

This correction is necessary to compensate for the different volume of reading and the re adsorption difference

of wire moistened with caprylic alcohol Open cock C Slowly *open* cock D and allow the 1 5 ml of acid to flow into the chamber *without allowing any air to enter* Close cock D and place a few drops

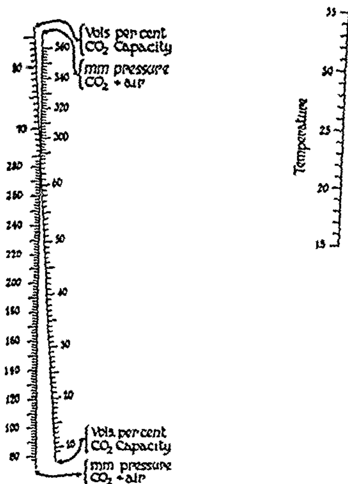


FIG 57 *Nomogram* for calculating CO_2 combining capacity of Van Slyke and Cullen manometric readings A straight line intersecting the temperature and mm pressure CO_2 + air scales at points of observed values cuts the vol per cent CO_2 capacity scale at the point showing the CO_2 capacity of the plasma when the plasma sample analysed is 1 ml (?)

The burette C is connected by rubber tubing to a levelling tube F which also contains water plus trace of sodium taurocholate At the start of an analysis the two water menisci are brought to a levelling mark on tubes X and Y by raising or lowering the levelling tube D while the 3 way taps are open to air The taps are now turned so that the bottles A and B communicate only with X and Y but are shut off from the air and the analysis is begun

If during the course of the analysis the volume of gas in A is altered by a change of external temperature or barometric pressure a similar change will occur in the compensator bottle B and the meniscus in Y will be displaced from the mark this change can be compensated by suitable movement of the levelling tube D and as this levelling tube communicates with X as well as with Y the compensation will also be applied through X to the bottle A provided that the meniscus in X is also brought back exactly to its mark by adjustment of E If these precautions are taken every time a reading of the burette has to be made all the readings will be strictly comparable with one another without further corrections for changes of external temperature or pressure

The whole apparatus (including bottles A and B) is immersed to the level of the taps in a stirred glass fronted tank of water maintained at a constant uniform temperature To keep the water tank free of moulds the use of methyl *p* hydroxy benzoate ½ oz to a gallon is advised

Reagents (1) *Borate solution* Add 12 404 g of pure dry boric acid to 100 ml of N caustic soda and dilute to 1 litre

(2) *Borate buffer pH* 10 Add 300 ml of the above solution to 200 ml 0 1N caustic soda

(3) *Saponin ferricyanide solution* Dissolve 0 3 g of saponin of known high lytic power and 0 6 g of potassium ferricyanide in 10 ml of water

Method

Oxygen Unsaturation

Place in bottle A 3 ml of buffer solution and cool in the tank 3 ml of blood which has been collected into heparin under paraffin is drawn up into an Ostwald Van Slyke pipette and carefully delivered under the buffer The amount of blood used is determined by the degree of unsaturation of the sample for the

Blood gas Analysis by Haldane's Method
Afte Douglas & Priestley (6)

The general form of the apparatus is shown in Fig 58 The gases liberated from or taken up by the blood in the small thin walled bottle A which is connected to the rest of the apparatus by narrow bore rubber pressure tubing are measured in the burette C (a 1 ml measuring pipette graduated to 0 01 ml and capable of being read to 0 001 ml) The second thin walled glass bottle B with its *connecting tube having the same volume as the analytical bottle* and connecting tube serves as a thermo barometer to allow for

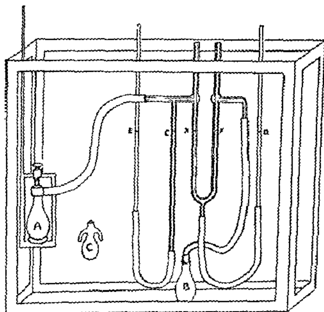


Fig 58 Haldane Apparatus

compensation of changes of temperature during the course of an experiment and should contain about 2 ml of water to ensure that the air in it is always saturated with aqueous vapour The two bottles A and B are connected through 3 way taps to the gauge tubes X *and* Y *respectively* which unite with one another below and are in communication with a levelling tube D X Y and D are partly filled with water *containing a trace of sodium tauro* cholate to lower the surface tension and ensure free movement

Reagents 1 0 05N *Caustic soda*

2 *Phenolphthalein* A 0 25% alcoholic solution

3 *Sodium biborate* A 2% aqueous solution

4 *Amino acid reagent* A 0 5% aqueous solution of sodium beta naphthoquinone 4 sulphonate prepared within five minutes of using

5 *Acetate buffer solution* 100 ml 50% acetic acid + 100 ml 5% sodium acetate

6 *Sodium thiosulphate* A 4% aqueous solution

7 *Stock standard* Take 536 mg of glycine dried to constant weight over sulphuric acid *in vacuo* and dissolve in 0 07N hydro chloric acid containing 0 2% sodium benzoate and make up to 1 litre with this solution 1 ml = 0 1 mg nitrogen

8 *Working standard* Dilute 2 ml of the stock standard to 50 ml with distilled water

9 *Dilute tungstic acid solution* Take 4 ml 10% sodium tung state + 4 ml 2/3N sulphuric acid and dilute to 100 ml with dis tilled water (This solution must be prepared and used within three hours)

Method To 6 ml dilute tungstic acid in a 15 ml centrifuge tube add 0 2 ml blood Allow to stand for five minutes and centrifuge Into a graduated tube place 5 ml filtrate and into another tube 5 ml water for blank Add 1 drop phenolphthalein to each tube and neutralise to a faint pink colour with 0 05N caustic soda To each tube add 1 ml 2% sodium borate and 0 4 ml of amino acid reagent mix and place in a boiling water bath for exactly three minutes Cool the tubes immediately in running water and add to them 1 ml of acetate buffer and 1 ml 4% thiosulphate Dilute to the 10 ml mark with distilled water and compare the colour of the unknown with the colour produced from the range of standards described below setting to zero with the reagent blank and using a blue filter maximum transmission 450 mμ The reading of the standard taken should be that nearest the reading of the unknown Prepare standards taking 1 2 3 4 and 5 ml of working standard making up in each case to 5 ml with water and treating as the 5 ml of filtrate above After experience with the method it is sufficient to use only the standards prepared from 2 and 5 ml

brighter red less unsaturated samples (*e g* arterial blood) 4 or 5 ml should be used Connect the bottle to the apparatus with the taps open to the air immerse adjust the menisci in tubes X and Y to the marks the pipette reading to about 0 7 ml and close the taps If the apparatus is suitably designed flush it through with carbon dioxide free air to reduce the time necessary for equilibrium by obviating absorption of carbon dioxide from the gas by the buffer Allow to stand when no further change in the pipette reading is noted return the menisci to their marks and read the pipette Shake bottle A to oxygenate the blood complete saturation has occurred when no further change is registered on the pipette or the levelling tube X which usually takes about 10 minutes Readjust the menisci X and Y and take pipette reading

Calculation

$$\frac{\text{ml } O_2 \text{ absorbed}}{\text{ml blood used}} \times 100 \text{ (correct to S T P)} = \text{ml oxygen absorbed}$$
per 100 ml blood

Oxygen Capacity

Oxygenate the blood sample provided by rotating it in a separating funnel Place into bottle C 2 ml of buffer followed by 1 ml of blood Into the side arm of bottle C place 0 5 ml of saponin ferricyanide solution Adjust the menisci as for determination of unsaturation except that the fluid level in the pipette is set at 0 3 ml Close the taps and shake the bottle until equilibrium is reached taking care not to mix the contents of the bottle with the side arm Adjust the menisci X and Y and note pipette reading a Tilt the bottle to run the saponin ferricyanide solution into the blood buffer mixture Shake again to expel all the oxygen from the blood and until a steady reading is obtained on the pipette and this usually takes about ten minutes readjust the menisci X and Y and take the pipette reading b

Calculation $(b - a) \times 100$ (Correct to S T P) = ml O_2 capacity per 100 ml blood

Some Colorimetric Methods

Estimation of Amino Acids in Blood

Adapted from the methods of Kravel (7) and Wilson & Fyles (8)

Method Into a 15 ml conical centrifuge tube place 5 ml of serum and 1 ml of 5N hydrochloric acid stir with a clean glass rod and allow to stand for about fifteen minutes Add 1 ml of sodium acetate followed by 3 ml of 25 / trichloroacetic acid shake and allow to stand for five minutes Place in a gently boiling water bath for about one minute to complete precipitation of the protein Cool and centrifuge at about 3 000 r p m for half an hour

Place 6 ml of *clear* supernatant fluid in a test tube followed by 0 1 ml of concentrated nitric acid shake and allow to stand for five minutes then add 2 ml of 5N potassium thiocyanate with gentle shaking

Blank To 5 ml of redistilled water in place of the serum add 1 0 ml of hydrochloric acid and proceed exactly as for the test

Reading Read blank and unknown in a null point colorimeter using a blue filter maximum transmission 500 mμ The unknown and the blank reading are converted to serum iron concentration by means of the calibration chart If the blank gives a reading greater than 25% of the total then the test is unsatisfactory because the reagents contain overmuch iron

The Estimation of Urinary Corticoids

Adapted from the method of Heard and Sobel (11) and Pincus (12)

Reagents Dilute immediately before use the *phosphomolybdic acid* reagent (described for blood sugar estimation on page 37_) with an equal volume of glacial acetic acid The precipitate that forms readily redissolves

Method Adjust to approximately pH 1 with concentrated hydrochloric acid a twenty four hour urine specimen collected without any preservative Stand the acidified urine at room temperature for twenty four hours to seventy two hours Extract the total sample four times in a separating funnel each time with a quarter of its volume of chloroform Combine all the chloroform extracts in a separating funnel and wash them with 3 × 100 ml of ice cold 0 1N caustic soda solution followed by washing with 3 × 100 ml of ice cold water Dry the washed chloroform extract by means of anhydrous sodium sulphate evaporate the washed dry chloroform extract to dryness in an all glass vacuum distillation apparatus keeping the temperature below 50 C and dissolve the gummy residue in 10 ml of dry chloroform Take an aliquot of this chloro

Calculation

1 ml standard = 2 5 mg amino acid nitrogen/100 ml blood
2 ml = 5 0 mg
3 ml = 7 5 mg
4 ml = 10 0 mg
5 ml = 12 5 mg

Blood amino acid concentration = Reading of unknown / Reading of standard × concentration of standard

Estimation of Serum Iron

Modified from Hoyer (9) and Dahl (10)

Reagents 1 *Water doubly redistilled* from glass

2 *Concentrated hydrochloric acid*

3 *Concentrated nitric acid* Reagents 2 and 3 are the constant boiling pure acids prepared by redistilling A R concentrated acids

4 *25% trichloroacetic acid* This is prepared from A R trichloroacetic acid further distilled using an all glass air cooled still and the redistilled acid dissolved in glass distilled water

5 *Approx 3N potassium thiocyanate solution* prepared by dissolving 10 g of A R thiocyanate in 20 ml of redistilled water This solution must be prepared on the day of the test

6 *Stock standard iron solution* (1 ml = 100 micrograms iron) 0 864 g of A R ferric ammonium sulphate is dissolved in about 100 ml of redistilled water 10 ml of pure hydrochloric acid (-) are added and the mixture is made up to a litre From this solution suitable dilutions can be made and a calibration curve constructed covering the range 0–400 micrograms of iron/100 ml

7 *Sodium acetate solution* A 20% solution of A R sodium acetate in redistilled water

All these reagents must be stored in pyrex bottles

Precautions The estimation is carried out on serum which has been collected with special care to avoid contamination with free iron at least 12 ml of blood are needed which should be taken with a dry all glass syringe using a new stainless steel needle The blood is allowed to clot and the serum separated a minimal amount of hemolysis does not interfere with the estimation All glassware used for the test must be specially cleaned (soaked in dichromate and finally rinsed with doubly distilled water) to ensure its being iron free

hydroxide in 10 ml of absolute alcohol using a mechanical shaker Filter through hardened filter paper (Whatman No 50 or 54) The solution should be crystal clear

7 *Meta dinitrobenzene* This material must be very pure The compound as supplied commercially is further purified as follows — Dissolve 20 g of *m* dinitrobenzene in 750 ml of 95 % alcohol and warm to 40 C Add 100 ml of 2N sodium hydroxide After five minutes cool the solution and add 2 500 ml of water Collect the precipitated *m* dinitrobenzene on a Buchner funnel wash thoroughly with water suck dry and recrystallize twice in succession from 120 ml and 80 ml of absolute alcohol by dissolving in the quantity of alcohol stated and evaporating off alcohol until crystallization commences

The purified material must be well crystallized almost colourless needles of melting point 90 5 to 91 C A mixture of a 1 / alcoholic solution of the compound with an equal volume of aqueous 2N caustic soda should give no colour after an hour

Use a 2 5 / solution of purified *m*-dinitrobenzene in absolute alcohol as the reagent Store in a dark bottle at 0 C This solution keeps for approximately one month but each time before use the excess solid should be brought into solution by gentle warming

Method *Hydrolysis and extraction* Measure the urine volume Into a 500 ml round bottomed flask place an aliquot of the mixed specimen (100 ml is suitable) add 15 ml of concentrated hydrochloric acid and 50 ml benzene Boil in a 500 ml ground glass flask for thirty minutes under a reflux condenser Cool fairly rapidly Transfer to a separating funnel withdraw the benzene and re extract the aqueous phase with two further 50 ml amounts of benzene

Combine the three benzene extracts and transfer to a separating funnel and wash consecutively with 2 × 25 ml of N sodium hydroxide followed by washing with 2 × 25 ml of N/2 hydrochloric acid and finally washing with 3 × 25 ml of water Evaporate off the benzene from the washed extract by distilling on a water bath under reduced pressure By means of about 20 ml of petrol ether transfer the ketosteroid extract quantitatively to a test tube making several rinses (the petrol ether insoluble material is non steroid pigment) Carefully evaporate to dryness in a water bath and dissolve the residue in 1 ml of *absolute* alcohol

form solution (containing 0 01 to 0 1 mg of corticoid material) and evaporate to dryness in a test tube (0 2 ml is usually a convenient amount) Dissolve it in 0 1 ml of glacial acetic acid and use another 0 1 ml of acid in another tube for the blank To each add 2 ml of phosphomolybdic acid reagent Plug the tubes very tightly with cotton wool wrapped in filter paper to prevent ingress of water vapour and immerse the lower third of the tubes in a gently boiling water bath for sixty minutes Cool the tubes for two minutes in running water and dilute each with a further 8 ml of phosphomolybdic acid reagent mix and allow to stand for two minutes for gas bubbles to rise

The colour density is read on a null point colorimeter against the blank determination (which should be colourless to the naked eye if the reagents are pure) using red filters with maximum transmission at 650 mμ

The colorimeter reading is converted to mg of corticoid present in the tube by means of a calibration curve

A calibration curve is prepared using as the standard material 0 01 to 0 1 mg of deoxycorticosterone in suitable steps *This method gives a straight line calibration curve so it is legitimate* and convenient to use a factor multiplying the colorimeter reading to give mg deoxycorticosterone equivalent to the corticoids present in the estimation tube

NOTE —This method although giving consistent figures is not now considered to estimate the true urinary corticoid content

Estimation of Total Neutral 17 Ketosteroids

Adapted for rapid clinical estimations from the method of Talbot et al (13)

Reagents 1 *Sodium hydroxide solutions* (*a*) 1N *solution* (*b*) 2 5N *solution*

2 *N/2 hydrochloric acid*

3 *Absolute ethyl alcohol* Aldehyde content must be lower than 0 0025 /

4 *Diethyl ether* Peroxide free—A R quality

5 *Benzene* (crystallizable)

6 2 5N *potassium hydroxide* in absolute ethyl alcohol solution The solution is made fresh daily by dissolving 1 4 g A R potassium

acidified urine add 1 ml of 20% sodium acetate and one drop of 0 02% alcoholic methyl red Add strong ammonia until just alkaline then acidify with glacial acetic acid and add 1 ml of ammonium oxalate solution After allowing to stand centrifuge Discard the supernatant fluid and dissolve the precipitate in 1 ml of normal hydrochloric acid Titrate at 60 C with decinormal potassium permanganate until the solution has a distinct pink colour Add one drop of 3% hydrogen peroxide 1 ml of sodium acetate solution and 1 ml ammonium oxalate solution Centrifuge wash the precipitate and titrate exactly as for the serum calcium

The *ionised calcium* (Maclean and Hastings (16)) is obtained by estimating the serum total calcium as given by the method of Clark and Collip (15) and the serum total protein by any suitable method

Knowing both of these values the percentage of ionised calcium can be read off the nomogram shown (see Fig 59)

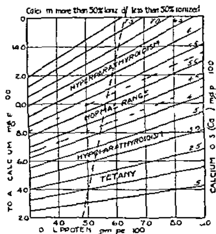


Fig 59 Chart for calculation of Ca++ concentration from total protein and total calcium of serum or plasma

Serum *carotenoids* are estimated by the method of Connor (17) For this method a working standard curve is prepared from potassium dichromate calibrated by comparison with pure β carotene (obtainable from the National Institute of Medical Research)

Select two glass stoppered tubes of approximately 15 ml capacity and pipette in respectively 0 2 ml of alcoholic solution and 0 2 ml alcohol for the blank Into each tube pipette 0 2 ml of 2 5% alcoholic *m* dinitrobenzene solution and 0 2 ml of 2 5N alcoholic potassium hydroxide solution Mix and place the tubes in a water bath at 25 C for one hour Add 10 ml of absolute alcohol to each tube Mix and read in a null point colorimeter within five minutes of dilution using yellow green filters of maximum trans mission 550 mμ If the unknown gives an optical density reading above the range of the calibration curve repeat the colour develop ment using a smaller quantity of alcoholic solution

A calibration curve is prepared using as the standard material *iso dehydro androsterone* *For calibration purposes a suitable* range to construct is from zero to 0 4 mg of sterone per 0 2 ml of alcohol

Patterson Reaction for Iso Dehydro Androsterone

Method of Patterson (14)

Evaporate to dryness 0 2 ml of the neutral alcohol extract obtained from the estimation of 17 ketosteroids Add 1 ml of concentrated sulphuric acid and shake until the steroid gum completely dissolves Place into a water bath at 25 C for twenty minutes Cool by immersing in cold water and add 1 ml of distilled water drop by drop with constant shaking Heat the mixture in a boiling water bath for one minute during which time a blue colour develops if the test is positive

Other Routine Analytical Methods

In this section are listed the methods necessary for the clinical biochemistry laboratory which are not fully described above and which comply with the essential requirements of routine analysis (page 350) Useful modifications of the standard procedures are described under the appropriate headings

The method generally used for the estimation of serum *calcium* is that of Clarke and Collip (15) (A) This involves the precipitation of calcium as an oxalate which is suitably washed and titrated with potassium permanganate It can be adapted for the estimation of urinary *calcium* provided a double precipi tation technique is used For this purpose take ₓ ml of slightly

20 30 10 50 60 70 80 and 90% and these after being made alkaline read on a null point colorimeter

For the *galactose tolerance test* Maclagan (27) has described a method of estimating the blood galactose after glucose has been removed by yeast fermentation by a modification of the Schaffer Hartman blood sugar technique

Para aminosalicylic acid in serum may be estimated by the method of Tarnoky and Brews (28) the *p* aminosalicylic acid is decarboxylated and the *m* aminophenol estimated by a diazotisation procedure

Numerous methods are available for the estimation of the serum phosphatases For *acid phosphatase* a standard procedure is that of Gutman and Gutman (29) (A) and for *alkaline phosphatase* that of King et al (30) (A) is probably the best method

Quinaldine red is a reagent which has recently been proposed for the estimation of *inorganic phosphorus* by Soyenkoff (31) A method using this reagent which forms an insoluble dye phos phomolybdate complex which remains in suspension can be used with very small quantities of blood (or other material) to give very reliable results

The standard method for the estimation of *plasma proteins* is by means of the Kjeldahl technique of nitrogen assay (C) This procedure has been used for many years and is probably still the most accurate Its main disadvantage is the limitation of the number of analyses which may be performed at the same time and other procedures such as the method of Kingsley (3°) (B) based on the biuret reaction have been proposed The biuret methods enable more analyses to be made at the same time but these methods do not possess the same degree of accuracy as does the Kjeldahl method The fractionation of the proteins is usually achieved by means of sodium sulphate sodium sulphite or ammonium sulphate and most laboratories standardise on one procedure and have their own normal values although such salting out methods do not necessarily agree with the pattern of the plasma proteins given by electrophoresis

Aggeler et al. (33) and Biggs and MacFarlane (34) have proposed methods for the estimation of *prothrombin time and concentration* The essential point of these methods is the preparation of the brain thromboplastin as a fine granulated powder This is done

To estimate *chlorides* in urine blood or cerebro spinal fluid the method of Van Slyke and Hiller (18) (B) is recommended The advantage of this method is that it can be adapted for the analysis of as little as 0 04 ml of material

Serum or plasma *cholesterol* is best estimated by a method based on the original technique of Bloor (19) such as that of Schoenheimer and Sperry (20) (B) The disadvantage in the use of these methods is that they tend to give higher results than those in which the material to be extracted is first absorbed on to sodium sulphate or filter paper The only points of technique worth noting are that water must be excluded during the development of the colour and any filter papers used must first be rendered lipoid free by extraction with chloroform

For the estimation of *creatine* and *creatinine* the best method is that of Peters (21) This method may also be adapted for urine estimations by suitable dilution of the urine based upon its specific gravity dilute the urine 4 × the last two figures of the specific gravity (e g S G 1006 dilute twenty four times) and the urine is then treated exactly as for the blood method

The flocculation tests recommended for investigating liver function are the *cephalin cholesterol colloidal gold* and *thymol turbidity* For the thymol turbidity and colloidal gold the methods of Maclagan (22 and 23) are preferred In the cephalin cholesterol test the method of Steinberg (24) is most satisfactory as a chemical antigen is used which is very stable and can be kept for months without deterioration This antigen consists of a mixture of pure desoxycholic acid and cholesterol

The serum *bilirubin* may be estimated by several different methods none of which are absolutely accurate a satisfactory method is that of King and Coxon (25)

The estimation of serum *bromsulphthalein* is best achieved by the method of Rosenthal and White (26) As we find that the colour of permanent alkalised bromsulphthalein tends to fade it is advisable to prepare a calibration curve The standard curve is constructed by taking a pooled normal sample of serum which has bromsulphthalein added to it to a concentration of 10 mg per 100 ml This concentration is considered as 100% and suitable dilutions are then made with more serum to obtain concentrations equivalent to 10

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Aggeler et al (33) and Biggs and MacFarlane (34) have proposed methods for the estimation of *prothrombin time and concentration* The essential point of these methods is the preparation of the brain thromboplastin as a fine granulated powder This is done

by macerating fresh human brain from the post mortem room (or rabbit brain if human is not obtainable) with acetone and repeatedly extracting with further quantities of acetone and finally drying at 37 C *The powder is stored in small quantities* in an evacuated desiccator over calcium chloride in a refrigerator It is standardised by testing against at least a dozen normal plasma specimens and determining the necessary quantity of brain powder and concentration of calcium ion to yield a prothrombin time in *normal plasma between fourteen and sixteen seconds A standard* curve is compiled by pooling the normal samples which fall within this standard range and diluting the (theoretically 100% prothrombin) pooled batch with physiological saline to contain 50 30 20 10 and 5 % of plasma then plotting the graph of prothrombin time against concentration *This graph must be reconstructed* for each fresh batch of thromboplastin and also checked monthly against known normals

The old and well tried method of Folin and Wu (35) (B) is still one of the best for the routine estimation of *blood sugar* A modification which may be made is that after the final colour has been developed with phosphotungstic acid the mixture should be again heated in the boiling water bath for three minutes then cooled diluted and read in a colorimeter This re heating technique renders the coloured material more stable over the period of reading The same method may be used for the estimation of *urinary sugar* after the urine has been diluted to bring the concentration of sugar to between approximately 0 1 / and 0 2% *It is also advisable to* remove interfering substances from the urine before dilution this is best done by adding to 1 ml of the urine 1 ml N/10 sulphuric acid 0 2 g of Lloyd s alkaloidal reagent 1 g of permutit and 8 ml of water in a stoppered tube vigorously shaking for five minutes filtering and diluting the filtrate as necessary

The Barrett urease method (36) (A) for the estimation of *blood urea* is satisfactory and his recommended technique of adding a few drops of 10 % sodium hypochlorite solution to the Nessler s solution (Koch & McMeakin (37)) is well worth the extra time *involved* the modification has the advantage that any trace of organic material present in the original solution does not interfere and cause a cloudiness of the final coloured solution The same method may be adapted for the estimation of *urinary urea*

dilution of the urine being effected by a calculation based on the specific gravity 1 ml of urine is diluted to 5 10 25 or 50 ml the final volume being the figure nearest the last two figures of the specific gravity *e g* Urine S G 10 1 dilute 1 ml of urine to 25 ml

This method of diluting urine may be used for numerous estimations such as *urinary phosphate urea or uric acid*

Apart from slight modifications no real advances have been made on the method of Folin (38) (B) for the estimation of *uric acid* and for clinical use this method is still quite satisfactory If used for urine it is first necessary to employ the silver precipitation technique (38) (B) where the urine is buffered silver nitrate added and the precipitate redissolved in the urea cyanide reagent

The components of cerebro-spinal fluid which are estimated as a routine are chloride sugar and protein The chloride may be assayed by the same method as already outlined for serum chloride namely that of Van Slyke and Hiller (18) (B) although some laboratories prefer to titrate the chloride direct with silver nitrate using dichlorofluorescein as the indicator (Rose (39)) Sugar is estimated exactly as for blood sugar whilst the protein level is obtained by the method of King and Haslewood (40) a method also of use for *urinary protein* estimation. The standards necessary for these protein methods may be purchased commercially

For the routine estimation of *fæcal fats* the method of Cambridge (41) still gives satisfaction although a more accurate estimation may be obtained by one of the methods where a prolonged Soxhlet extraction is employed (Harrison (C))

Estimations of *fæcal trypsin* are based on the digestion of different substrates by means of the enzyme in alkaline solution Anderson and Early s method (42) relies upon the liquefaction of a standard amount of gelatin by varying the amount of fæces mixed with sodium bicarbonate solution The result is expressed in units representing the reciprocal of the limiting dilution necessary to digest a given amount of gelatin *e g* limiting dilution 1 *g* of fæces/100 *g* of sodium bicarbonate solution—result 100 units/*g* wet fæces It should be noted that this analysis may only be carried out on fresh fæces as trypsin rapidly decomposes The method may also be used for the assay of *trypsin in duodenal juice*

The estimation of *fæcal urobilinogen* is now coming into more

general use and a suitable method for its estimation is that of Maclagan (43) In this method an artificial standard is used consisting of a alkaline solution of phenolphthalein and this is compared with an extract of feces which has been treated with alkaline ferrous sulphate to reduce any urobilin present to urobilinogen before reacting it with Erhlich s aldehyde reagent

For the routine clinical estimation of *urinary ascorbic acid* the use of 2 6 dichloroindophenol is still the simplest technique Harris and Ray (44) have produced a method based on this reaction but if a more accurate estimation is required or if it is desired to estimate the ascorbic acid in blood use should be made of one of the methods where the ascorbic acid is reacted with 2 4 dinitrophenol hydrazine (Roe and Kuether (45))

When estimating *urinary diastase* by the method of Wohlgemuth (46) it is essential that the urine should first be suitably buffered at pH 6 1 with phosphate buffer (B page 312)

The estimation of *urinary galactose* may only be required for the galactose tolerance test The method for blood galactose is satisfactory after suitable dilution

After the intravenous injection of sodium benzoate where it is desired to estimate the excretion of *hippuric acid* the method of Quick et al (47) is eminently suitable The most difficult point in this technique is to obtain the crystals of hippuric acid from the concentrated acidified urine Friction is the best method to initiate their formation from the urine after it has once been cooled although in cases of extreme difficulty it is permissible to seed with a very small crystal of pure hippuric acid

Urinary pregnanediol is best estimated by slight modification of the method of Somerville and Marrian (48) where the urine is hydrolysed by boiling with hydrochloric acid and extracting with toluene as one operation The toluene is washed with caustic soda to remove impurities evaporated under dryness and the pregnanediol precipitated from an alcoholic solution of the gum under carefully controlled conditions with caustic soda The precipitated material is then reacted with concentrated sulphuric acid to give the yellow colour by which it is estimated in comparison with a previously constructed calibration curve prepared from pure pregnane 3 α 20 α diol

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